Physiological Phenotyping of Fronto-temporal lobar degenerations

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A Thesis

Presented for the Degree of

Doctor of Philosophy

The Dementia Research Centre

Institute of Neurology

University College London

June, 2015

Declaration statement

I, Phillip Fletcher, confirm that the work presented in this thesis is my own. Where information has been derived from other sources, I confirm that this has been indicated in the thesis.

Phillip Fletcher 10th June 2015

Summary

Fronto-temporal lobe degenerations encompass a spectrum of neurodegenerative diseases underpinned by pathological protein deposition. The study of these diseases is of considerable interest, both from a neurobiological perspective of brain organization in health and disease, and in order to develop novel biomarkers. However, the translation of the effects of abnormal protein to clinical syndrome is far from clear and whilst the current classification systems provide a framework upon which to base evaluations, they do not capture the full spectrum of the complexity of these disease entities. Recently there has been a shift in collective thinking towards describing brain functions in terms of the activity of inter-connected neuronal 'networks' rather than as the discrete functional areas, networks that underlie core physiological processing systems. There are symptoms in FTD that, whilst not the most prominent, may speak to derangement of these physiological systems, systems involved in processes such as salience, hedonic and emotion appraisal. Here I propose that using a physiological approach to examine these symptoms may allow insight into the real time in-vivo physiological effects of proteinopathies upon dynamic neuronal systems. Five studies are presented investigating FTD in relation to AD and healthy older adults. In experiment 1 behaviours suggestive of abnormalities of pain and temperature perception are investigated to both better characterize the nature of these symptoms and investigate underlying neuroanatomical correlates. In experiment 2, 3 and 4 the physiological effects, as measured by pupillometry, of manipulations of three levels of salience cues are examined and in the final experiment the effects of hedonic and emotion processing from sound and music are explored. Taken together these experiments provide insight into disordered physiological processing in these diseases and offer new metrics for a physiological approach to the assessment of these patients.

Contents

Abbreviations

ACC: anterior cingulate cortex
AchEI: acetylcholine esterase inhibitor
AD: Alzheimer's Disease
ALS: amytrophic lateral sclerosis
Anti-d: anti-depressant
ATL: anterior temporal lobe
Arith: arithmetic
BP: blood pressure
BPVS: british picture vocabulary score
bvFTD: behavioural variant fronto-temporal Dementia
C9orf72: chromosome 9 open reading frame 72
CBD: corticobasal disease
CBS: corticobasal syndrome
CSF: cerebrospinal fluid
dB: decibels
dd: disease duration
DMN: default mode network
DKEF: Dellis-Kaplan executive function
dACC dorsal anterior cingulate cortex
DTI: diffusion tensor imaging
Edc: education
fMRI: functional magnetic resonance imaging
FTD: fronto-temporal dementia
FTLD: fronto-temporal lobe degeneration
FUS: fused in sarcoma protein
GNT: graded naming test
GRN: progranulin
HADs: History of anxiety and depression scale
HC: healthy controls
HR: heart rate
IQ: intelligence quotient
MAPT: micro-tubuole associated protein tau
MMSE: mini mental state examination
MNI: Montreal neurological institute
MRI: magnetic resonance imaging
OFC: orbitofrontal cortex
PET: positron emission topography
PNFA: progressive non-fluent aphasia
PIQ: performance intelligence quotient
PPA: primary progressive aphasia
PSP: progressive supranuclear palsy
R ALT: right anterior temporal lobe
RMS: root mean square
RMT: recognition memory test
SCR: skin conductance response
SD: semantic dementia
SMT: semantic matching test

SPM: Statistical parametric map
STS: superior temporal sulcus
Syn: synonym test score
TDP: TAR DNA-binding protein-43
TPJ: temporo-parietal junction
VBM: voxel based morphometry
VIQ: verbal intelligence quotient
VOSP: visual object and space perception battery
VTA: ventral tegmental area
WASI: Wechsler abbreviated scale of intelligence

Figures and Tables

FIGURES:

TABLES:

Chapter 1. Introduction

1.1 What is Fronto-temporal Dementia?

Fronto-temporal lobe degeneration (FTLD)[1] is a generic term that encompasses a spectrum of clinically, pathologically and genetically heterogeneous neurodegenerative diseases. FTLD is the second commonest cause of young onset dementia after AD and the point prevalence at has recently been estimated at approximately 15-22/100,000, with an incidence of 2.7-4.1/100,000 (Onyike & Diehl-Schmid, 2013). Symptoms usually develop in the sixth decade of life, but onset is highly variable (Neary *et al.*, 2005; Warren *et al.*, 2013a). As the name implies, FTLD is associated with frontal and temporal lobe atrophy, with relative preservation of posterior cortical and memory functions, at least early in the disease (Hornberger & Piguet, 2012). The hallmark clinical features of the FTLD syndromes are a progressive and insidious decline in cognition underpinned by the deposition of a range of abnormally folded proteins (Graveland *et al.*, 1985; Seeley *et al.*, 2006). Based upon the leading symptom, the FTLD syndromes are nosologically divided into a behavioural (bvFTD) and two language led variants; semantic dementia (SD) and progressive non-fluent aphasia (PNFA) (Rascovsky *et al.*, 2007; Gorno-Tempini *et al.*, 2011). The bvFTD syndrome is led by progressive behavioural decline with an insidious destruction of personality, social conduct, and social cognition (Rascovsky *et al.*, 2007; Hornberger *et al.*, 2010a). SD is primarily a disorder of the semantic system, initially language led (Hodges & Patterson, 2007), but followed by a generalised sensory object agnosia with semantic impairment demonstrated in the visual, auditory, tactile and chemo-sensory domains demonstrated (Bozeat *et al.*, 2002; Rami *et al.*, 2007; Goll *et al.*, 2010; Piwnica-Worms *et al.*, 2010). PNFA is primarily a progressive disorder of motor speech output (apraxia of speech) and agrammatism (Gorno-Tempini *et al.*, 2004; Ash *et al.*, 2010). Diagnostic criteria for

[1] The term FTLD is used here to denote pathological demarcation of diseases and FTD when referring to clinically defined syndromes.

each clinical syndrome can be found in Appendix 10.1.

Around a quarter of FTD is familial (Kirshner, 2014). However, despite considerable recent interest, mechanisms by which these genetic and protein abnormalities map onto clinical phenotype remain enigmatic with different phenotypes arising from a given protein or genetic abnormality, and conversely, the same clinical picture underpinned by a range of molecular abnormalities. Further, despite the segregation of a spectrum of disease phenotype into three canonical syndromes, there remain symptoms that traverse these divisions that, although little studied, may speak to derangements of more widespread core processes basic to biological drives, such as those involved in salience, positive reinforcer and emotion evaluation. One potential avenue to unify what may currently appear to be a heterogeneous collection of disorders may be not to consider neurodegenerative diseases as diffuse processes spreading confluently from a stochastic insult, but to think more in terms of the effects of abnormal proteins upon the vulnerable neuronal networks underpinning key biological systems (Seeley *et al.*, 2009; Fletcher & Warren, 2011). Evaluation of disordered processing within these systems may not only shed light upon normal processing of core biological drives, but allow insights into disease mechanisms and the development of novel in-vivo metrics of disease.

Amnestic AD is also included in this Thesis as a disease control group in which to compare and contrast effect demonstrated in FTD. Like FTD, AD is a neurodegenerative disorder associated with the deposition of mis-folded protein aggregates. An amnestic presentation indicative of underlying hippocampal dysfunction accounts for the majority of cases (Mahoney *et al.*, 2011). In the early stages these deficits may remain focal (Moss *et al.*, 1986; Cohen *et al.*, 1997), but inevitably insidiously spread, with the emergence of temporo-parietal syndromes (McKhann *et al.*, 1984; Storey *et al.*, 2002; Mahoney *et al.*, 2011), working memory (Rochon *et al.*, 2000), visual object perception (Fujimori *et al.*, 1997; Adduri & Marotta, 2009), and semantic memory deficits occurring (Chertkow & Bub, 1990; Greene & Hodges,

1996).

1.2 Pathology:

The initial FTLD cases described by Alois Alzheimer displayed intraneuronal inclusions upon histopathological examination, termed 'Pick bodies'[2]. These inclusions were later found to also stain for the protein microtubule-associated protein tau (MAPT) (Buee & Delacourte, 1999) and it has subsequently been demonstrated that in fact only a small number of cases of FTD are associated with pick bodies (Snowden *et al.*, 2002). With refinement of histopathological staining techniques a classification system based upon staining for three major proteins, microtubule-associated protein tau (FTLD-TAU), TAR DNA-binding protein-43 (FTLD-TDP), and fused in sarcoma protein (FTLD-FUS) is now widely used (Mackenzie *et al.*, 2010; Seelaar *et al.*, 2011). Of these three 'proteinopathies', TDP-43 is the most common, accounting for around 50% of FTD cases, with tau underpinning for around 45% and FUS 5%, of cases (Rohrer & Warren, 2011). Around a quarter of FTLD is familial, with the strongest genetic component found in the behavioural form (Kirshner, 2014). The most common mutations occur in three genes; progranulin (GRN), microtubule associated protein tau (MAPT) and the chromosome 9 open reading frame 72 (C9orf72) gene (Rohrer *et al.*, 2009; Rohrer & Warren, 2011). AD is characterized by two neuropathological hallmarks: extracellular deposits of amyloid-beta and intracellular neurofibrillary tangles (Braak & Braak, 1991).

[2] It is worth noting that it was upon this basis that the disease (most commonly clinically mapping to a behavioral form (see below)) was termed 'Pick's disease', which is still used by some authors synonymously with the behavioural variant form of FTD, bvFTD).

1.3 Proteinopathies in FTLD do not clearly predict clinical phenotype:

The relationship between abnormal protein and clinical phenotypes remains far from clear, with the same protein underpinning a variety of clinical syndromes (Cairns *et al.*, 2007; Weintraub & Mesulam, 2009); tau for example has also been shown in association with a phenotypically heterogeneous group of neurodegenerative disorders, referred to as 'tauopathies', that include syndromes such as progressive supranuclear palsy (PSP) and corticobasal syndrome (CBS) that are also considered part of the FTLD spectrum, but lie outside the canonical FTLD classification (Cairns *et al.*, 2007; Robert & Mathuranath, 2007). Conversely, the same clinical syndrome can be found in association with a variety of proteins, with a recent large study showing that although the majority of SD is underpinned by FTLD-TDP (68% case) and about half of PNFA and bvFTD cases are underpinned by tau (50% and 42% respectively), with the underlying pathology of the latter most heterogeneous (Chare *et al.*, 2014). All clinical phenotypes were additionally found to be associated with alternative FTLD pathologies (tau, FTD-TDP, FUS and 'FTLD-other') with 26% of cases secondary not to FTLD pathology at all, but to Alzheimer pathology (9% bvFTD, 16% SD, 31% PNFA) (Chare *et al.*, 2014). Four isoforms of TDP-43 have been described (Mackenzie *et al.*, 2011) which associate more (although incompletely) with particular clinical phenotypes; Type A is mainly associated with bvFTD and PNFA, Type B is found in association with bvFTD and FTD-MND, Type C is associated with SD and bvFTD and Type D is found in association with inclusion body myositis and Paget's Disease (Mackenzie *et al.*, 2010). The clearest mapping occurs between Type C and SD (Josephs *et al.*, 2011; Rohrer *et al.*, 2011).

A similar situation is found with the relationship between underlying genetic abnormality and clinical syndrome, as even when a genetic abnormality is known the associated clinical syndrome can be highly heterogeneous in both phenotype and natural history. For example, MAPT mutations, although most commonly associated with a bvFTD phenotype, have

also been found in association with parkinsonism and even a clinical picture and atrophy pattern mimicking amnestic AD (Liang *et al.*, 2014). GRN mutations are associated clinically with bvFTD, PNFA and also CBD, whilst expansions in the C9orf72 gene can result in phenotypes ranging from bvFTD to a pure amytrophic lateral sclerosis picture, PPA, or even amnestic AD (Harms *et al.*, 2013; Siuda *et al.*, 2014). Parkinsonism is variably seen with any of the mutations (Rohrer & Warren, 2011). For a given gene mutation kindred age of onset, clinical presentation and rate of progression can all vary; for example, within a known C9orf72 mutation family; age of disease onset has been shown to vary from 43-68 years old and disease duration to vary from 1.7-22 years (Downey *et al.*, 2013).

Therefore, even in cases where the likely underlying gene abnormality or likely proteinopathy can be predicted, the clinical phenotype that will arise is not necessarily clear, and conversely, the variability of potential underlying pathologies of a given clinical presentation, plus the clinical variability of subjects all within a particular phenotypic group, makes predicting the underlying protein abnormality from a clinical phenotype problematic and imprecise. Additionally, clinical syndromes often extend beyond the FTD spectrum, for example, co-occurrence with amytrophic lateral sclerosis (FTD-ALS), CBS and PSP are common (Rascovsky *et al.*, 2011; Mesulam *et al.*, 2012; Sajjadi *et al.*, 2012). In a recent study patients with pathologically confirmed diagnoses of FTD, all of whom had presented with a form of aphasia, were retrospectively clinically re-classified using current consensus criteria. Less than half (25 of 52) met diagnostic criteria with 22 displaying features from several diagnostic categories and 2 not meeting threshold for diagnosis (Harris *et al.*, 2013). Therefore, current methods of clinical classification do not capture the full spectrum of complexity of these disease entities. Clearly resolving this is important with the development of protein specific treatments on the horizon, where treatment successes and quantification of these will be confounded by pathological heterogeneity (D'Alton & Lewis, 2014).

The complexity of the mapping of abnormal protein to syndrome is illustrated schematically in Figure 1.1.

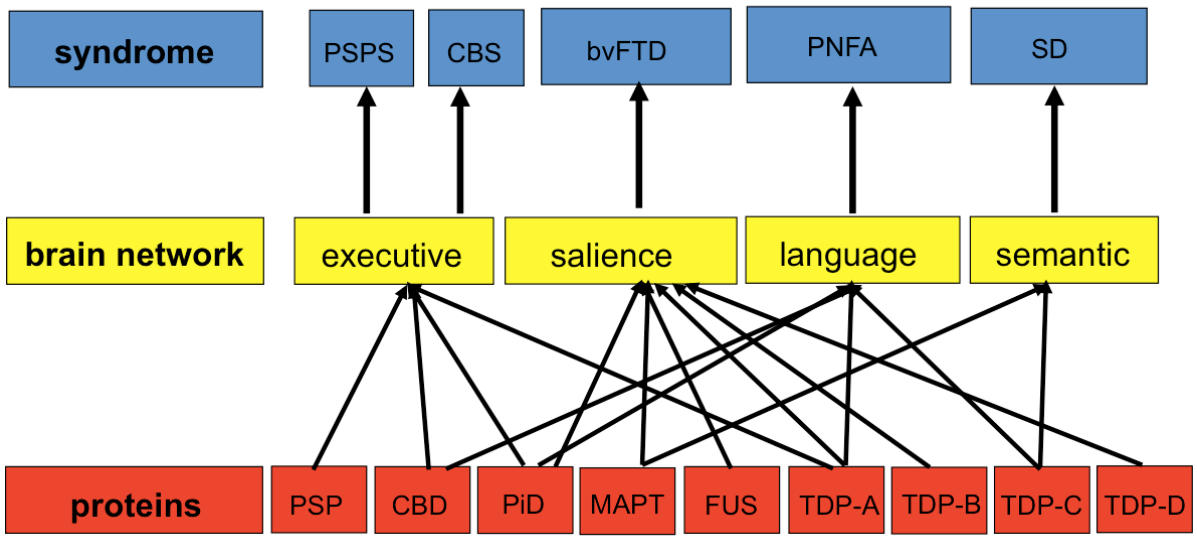

Figure 1.1 Schematic of the mapping of proteinopathy (red boxes) to the putative anatomical network affected (yellow boxes) and resultant clinical syndrome (blue boxes). The figure illustrated the idea that the same clinical syndrome can be underpinned by multiple protein abnormalities. Image adapted with permission from Jason Warren

1.4 A network perspective:

The difficulty in providing coherence between the underlying molecular substrates and the resultant clinical syndrome limits the potential to understand disease mechanisms or follow disease evolution. A shift in focus towards examining the effects of proteinopathies upon specific neuronal networks and the ensuing physiological system derangement may allow for better evaluation and is discussed below.

The three canonical FTD syndromes demonstrate characteristic profiles on neuroimaging, as measured by volume loss (atrophy) or decreased functional activity, relative

to healthy controls. In bvFTD, the frontal and temporal lobes, anterior cingulate cortex (ACC), ventral anterior Insula and subcortical structures such as the thalamus are the regions most consistently affected (Rosen *et al.*, 2002; Varrone *et al.*, 2002; Boccardi *et al.*, 2005; Schroeter *et al.*, 2007; Schroeter *et al.*, 2008; Richards *et al.*, 2009; Rohrer *et al.*, 2009). In SD the brunt of disease burden is borne by the dominant anterior-inferior temporal lobe and mesial temporal lobe structures, in particular the amygdala (Bathgate *et al.*, 2001; Gorno-Tempini *et al.*, 2004; Hodges & Patterson, 2007; Davies *et al.*, 2009; Rohrer *et al.*, 2009) (Whitwell *et al.*, 2005) and with disease progression the orbitofrontal cortex, insula and anterior cingulate become affected (Gorno-Tempini *et al.*, 2004; Rohrer *et al.*, 2009; Rogalski *et al.*, 2011). In PNFA damage maximally occurs in the left perisylvian fissure, frontal operculum, premotor and supplementary motor areas, and dorsal anterior insula (Josephs *et al.*, 2006) spreading with time caudally into the parietal lobe (Gorno-Tempini *et al.*, 2004; Schroeter *et al.*, 2007; Rohrer *et al.*, 2009; Goll *et al.*, 2010; Hu *et al.*, 2010). AD is associated with bilateral and symmetrical hippocampal atrophy that later incorporates parahippocampal regions, amygdala (albeit to a lesser extent than in SD) and the temporal lobes more widely, as well as the parietal and frontal lobes (Whitwell & Jack, 2005; Whitwell *et al.*, 2005).

Recently collective thinking has shifted towards describing brain functions in terms of the activity of inter-connected neuronal 'networks' rather than as discrete areas (Seeley *et al.*, 2009; Warren *et al.*, 2013b). In the healthy brain during task free conditions (i.e. in a 'resting state'), correlated spontaneous activity occurs in multiple discrete areas in temporally fluctuating and replicable patterns, now referred to widely as 'neuronal networks' (Greicius *et al.*, 2003; Fox *et al.*, 2005; Fransson, 2005; Fox & Raichle, 2007; Seeley *et al.*, 2007b) with different dementia syndromes showing abnormalities in spatially isolated regions that are part of the same network (Seeley *et al.*, 2007b). In healthy older adults, seeding connectivity analysis from those regions that are maximally atrophied in FTD and AD shows connectivity to a pattern of regions that are found to be atrophied in these diseases, providing support for the concept of

spread of pathological proteins through specific neuronal systems (Seeley *et al.*, 2009). However, the mechanisms by which pathological proteins target these specific networks remains to be established; spreading axonal degeneration secondary to axonal transport deficits of neuronal growth factors (Salehi *et al.*, 2006) and tran-synaptic direct spread of pathological proteins (Bartz *et al.*, 2002) are two possibilities. Indeed, abnormally folded tau has been shown to induce abnormal folding in nearby tau molecules (Frost *et al.*, 2009) and over time damage has been demonstrated to spread to new regions, regions that have known anatomical connections with sites of earlier insult (Seeley, 2008). Pathological proteins could potentially result in toxic loss or gain of function (Halliday *et al.*, 2012; Warren *et al.*, 2013b) with shorter range, clustered neurons appearing to be particularly vulnerable to some tauopathies (Ikeda *et al.*, 2005; McMillan *et al.*, 2008) whereas selective involvement of long range Von-Economo neurons would account for the often bi-hemispherically symmetrical atrophy patterns eventually observed even in relatively focal (at least in early disease) syndromes such as SD (Seeley *et al.*, 2006; Fletcher & Warren, 2011).

These networks differentially affected in FTD overlap those involved in a wide range of processes including attention, executive functions, appraisal, salience assignment and emotion processing, (Seeley *et al.*, 2007b; Guo *et al.*, 2013; Parks & Madden, 2013; Andrews-Hanna *et al.*, 2014), and evaluation of the effects of molecular abnormalities upon physiologically functions may allow observation of the real time in-vivo translation of the effects of pathological proteins on these networks. Better mapping of the functional consequences of diseases could allow for clearer stratification of phenotypes and more sensitive evaluation from a clinical perspective. This has far reaching implications: not only could this help enable better understanding of disease processes but this will also be important for tracking and monitoring progression and response to treatments within the context of treatment trials.

1.5 Physiology of sensory processing:

Whilst not the most prominent features and not included in the diagnostic criteria, deficits in emotion recognition have been demonstrated throughout the FTD clinical spectrum occurring in SD and, albeit to a lesser extent, in PNFA in addition to bvFTD (Rankin, Gorno-Tempini et al. 2006, Mendez and Shapira 2009, Kumfor, Miller et al. 2011, Rohrer and Warren 2011, Sturm, McCarthy et al. 2011, Rohrer, Sauter et al. 2012, Sturm, Yokoyama et al. 2013). These symptoms become more prominent as diseases progress reflecting an overlap of network dysfunction with time (Snowden, Bathgate et al. 2001, Rosen, Wilson et al. 2006, Bediou, Ryff et al. 2009, Rascovsky, Hodges et al. 2011, Kumfor and Piguet 2012, Perry, Sturm et al. 2014, Zhou and Seeley 2014). Conversely, increased emotional contagion has been demonstrated in AD (Sturm *et al.*, 2013). Due to the difficulties in characterizing and assessing these symptoms, they have received relatively little attention in the literature, however better evaluation may shed light upon the underlying pathological processes at work. Below I outline the hypothesis that behavioural deficits in emotional recognition observed in these patients reflect abnormalities in physiological processing on three main levels; firstly, derangement of brain encoding of the basic stimulus properties, secondly, disruption of the ability to appropriately proportion degree of salience and hedonic value to the stimuli, and finally, that deranged autonomic effector pathways result in abnormal behavioural responses and sensory feedback. The interactions of these factors together results in the clinical syndromes and offer different levels for physiologically probing these processes, with different disease groups, by virtue of seperable anatomical substrates, producing syndrome (or molecular abnormality) different 'physiological signatures'.

1.5.1 Abnormal sensory signal encoding:

Within the FTD literature, work into sensory coding has focused on investigation of alterations in the semantic analysis or valuation of sensory experiences, with patients with FTLD commonly failing to correctly recognise or interpret emotional and social cues (Jesso *et al.*, 2011; Omar *et al.*, 2011a; Kumfor & Piguet, 2012; Zhou & Seeley, 2014) and demonstrating obsessional attachment to particular stimuli such as sweet foods (Woolley *et al.*, 2014) or music (Fletcher *et al.*, 2013). This suggests a generic disturbance in processing of the hedonic value of stimuli (Perry *et al.*, 2014). However, several series have documented symptoms that might additionally reflect abnormal coding of somatosensory signals on a more basic level. Differential disruption in the encoding of incoming afferent sensory information has been demonstrated across sensory domains in FTD and AD with cross modal deficits of basic central perception (sound, in SD and PNFA and odour discrimination in PNFA and AD), and semantic processing (odour and flavour identification in bvFTD and SD) demonstrated (Rami *et al.*, 2007; Djordjevic *et al.*, 2008; Goll *et al.*, 2010; Piwnica-Worms *et al.*, 2010; Omar *et al.*, 2013; Heyanka *et al.*, 2014). Impairment of flavour identification has been shown to be associated with atrophy of the left entrorhinal cortex, hippocampus, parahippocampus and temporal pole in a combined FTD cohort (Omar *et al.*, 2013) whereas impaired odour identification has been demonstrated in association with atrophy of the right mid-frontal gyrus in bvFTD (Pardini *et al.*, 2009). Symptoms have also been reported that suggest distortions of the interpretation of somatosensory afferent information in FTD. Behaviours suggestive of abnormal pain and temperature perceptions are reported in both SD and bvFTD (Snowden *et al.*, 2001), as well as an array of poorly characterized visceral and somatic sensations occurring surprisingly commonly in around 40% of cases, which in some pre-date the onset of any diagnostic symptoms (Pijnenburg *et al.*, 2004; Landqvist Waldo *et al.*, 2014). Quantitative neuroanatomical correlates have not been made, although there appears to be a qualitative preponderance of abnormal sensory perceptions in conjunction with right anterior temporal lobe atrophy (as

opposed to left) within an SD cohort (Chan *et al.*, 2009). Whilst many of these symptoms could reflect abnormal assignment of salience (see below), it may be that these symptoms, like distortions of olfaction and sound processing, reflect more generally disrupted physiological sensory encoding systems on a basic processing level; if afferent sensory information cannot be accurately encoded then further evaluation will be inherently distorted.

1.5.2 **Abnormal evaluation of salience and reinforcement learning:**

The salience of a stimulus are those properties that allow it to stand out from the multisensory background and appropriate 'weighting' of the likely relevance of a stimulus for allocation of attentional resources is essential to an organism's survival; on the simplest level, those stimuli that signal danger are to be avoided and those that provide positive reinforcement in some way should be approached. A range of stimulus characteristics likely encode salience; for example, basic object properties (such as loudness in the auditory, or brightness in the visual, domains), and simple cues such as perceived direction of motion have been demonstrated to evoke orienting responses (Bach *et al.*, 2008; Wang *et al.*, 2012; Wang *et al.*, 2014; Wang & Munoz, 2014). However, two perceptually similar sources can have very different biological implications, and this is particularly apparent in the in the challenging realm of sound, (compare, for example, the rumble of thunder and the growl of a large predator). Therefore, salience is also likely influenced by semantic content and context, which has yet to be investigated. Equally, the more cognitively complex processes involving real world responses may be particularly salient; emotional cues for example, are among some of the most biologically salient, linked to basic biological drives relevant to social signalling, self-awareness and positive reinforcer seeking (Sturm *et al.*, 2006; Kumfor *et al.*, 2011; Chiong *et al.*, 2013; Sturm *et al.*, 2013; Perry *et al.*, 2014; Shany-Ur *et al.*, 2014). Therefore, one can hypothesise that many behaviours in FTLD and AD may reflect inappropriate assignment of the salience value of

emotional stimuli. This is probably most compelling from study of bvFTD patients where deficits in emotion recognition and expression (including more complex emotional behaviour such as empathy, humour or sarcasm recognition) are well demonstrated (Snowden *et al.*, 2003; Lough *et al.*, 2006; Rankin *et al.*, 2006; Kosmidis *et al.*, 2008; Hornberger *et al.*, 2009; Kipps *et al.*, 2009; Eslinger *et al.*, 2011; Omar *et al.*, 2011a).

In experimental paradigms investigating the neuroanatomical substrates of salience processing, a variety of methods have been employed to modulate the salience of experimental paradigms. This includes rendering previously neutral stimuli salient by associate learning (Roiser *et al.*, 2009), manipulation of the emotional dimensions of pain (Peyron *et al.*, 2000), empathy for pain (Singer *et al.*, 2004), metabolic stress, hunger or pleasurable touch (Craig, 2002), music (Blood & Zatorre, 2001) and social rejection (Eisenberger *et al.*, 2003); regions showing activation on fMRI include the ventral striatum, VTA, amygdala, prefrontal cortex, dACC and fronto-insula cortex.

Using resting state fMRI, the strength of the connections between the dACC and the fronto-insula cortex have been shown to correlate with out of scanner baseline self reported anxiety levels, and this has been postulated to indicate that those individuals with raised baseline anxiety states may have heightened salience processing systems (Seeley *et al.*, 2007b); these regions are key to a 'salience network' and act as key hubs in a system tasked with the assignment of degree of salience, irrespective of sensory modality. Whilst one must remain reticent about overstating the conclusions that can be drawn from the use of self-reported baseline anxiety levels as a proxy for relative salience (which after all only potentially reflect one aspect of salience processing), the fronto-insula cortex and dACC are regions consistently activated across studies and may represent key inter-connected hubs of a salience network (Downar *et al.*, 2000; 2001; 2002; Menon & Uddin, 2010), even if the exact boundaries of this processing system remain to be defined. This thesis does not aim to absolutely demarcate the

limits of this network, and the term 'salience network' is used throughout to refer to the concept of a brain processing system focused upon the fronto-insula cortex and ACC, concerned with the attribution of degree of salience of stimuli.

The ACC and fronto-insula are areas particularly bearing brunt of pathology in bvFTD (Seeley *et al.*, 2009) and it has therefore been proposed that behaviours key to FTD, such as apathy, emotional blunting and flattened affect, may reflect different aspects of a generally disrupted salience processing system and a failure to assign relevant salience to stimuli in general (Zhou & Seeley, 2014).

In Alzheimer's disease key areas affected mirror those that in the healthy brain have been shown to be functionally linked at rest and de-activated when engaged in various cognitive tasks, as part of a network termed the 'Default mode network' (DMN) (Shulman *et al.*, 1997; Raichle *et al.*, 2001). The exact roles and anatomical limits of this network are currently poorly defined, but in general are felt to include self-referential thought and self-reflection, episodic memory retrieval, mental state attribution, and visual imagery (Mason *et al.*, 2007; Buckner *et al.*, 2008) and in AD the strengths of these connections are attenuated, even in early clinical disease (Greicius *et al.*, 2004; Supekar *et al.*, 2008). In the healthy brain the DMN and salience network (fronto-insula cortex, ACC, dlPFC) have been demonstrated to show temporo-parietal junction (TPJ) mediated inversely correlated functional activity (Kucyi *et al.*, 2012a; Kucyi *et al.*, 2012b; Kucyi & Davis, 2014). Interestingly in bvFTD decreased salience network activity occurs in conjunction with increased DMN activity with the opposite pattern occurring in AD, and it is this relative imbalance of interaction between these two systems that has been proposed to account for the symptoms observed; whilst decreased SN function accounts for flattened emotional reactivity in bvFTD, increased salience network activity underpins the retained or enhanced warmth and empathy, sharing of emotional states, emotional morality and heightened emotional 'contagion' reported in AD (Rankin *et al.*, 2006; Mendez & Shapira, 2009; Sturm *et al.*,

2011; Sturm *et al.*, 2013).

Recent work has also suggested that disrupted of positive reinforcement learning may underlie some of the behaviours observed in bvFTD, such as the evolution of binge eating (abnormal eating behaviours occur in over 80% cases at some point in the disease (Hornberger *et al.*, 2009)), and the rarer symptoms of hypersexuality and new drug and alcohol use (Miller *et al.*, 1995; Cruz *et al.*, 2008; Mendez & Shapira, 2013). A positive reinforcer is something that produces a sense of pleasure, which in turn tends to lead to repetition of behaviour. Work with both animals and humans has repeatedly implicated a network of key cortical and subcortical brain regions in reinforcement behaviours, networks that overlap the salience network as defined by Seeley et al (2007). In animal studies exploring the positive reinforcing properties from eating (Hernandez & Hoebel, 1988), sex (Pfaus *et al.*, 1995) and administration of drugs of abuse (Carelli, 2002), the ventral striatum is consistently implicated (particularly the nucleus accumbens and ventral pallidum), in particular in conjunction with areas including the ventral tegmental area (VTA), amygdala, hippocampus, ventro-medial pre-frontal cortex, hypothalamus and dorsal mid-brain (Pfaus *et al.*, 1995; Bardo, 1998; Schilstrom *et al.*, 1998). A similar anatomical pattern is observed in humans, with the ventral striatum (particularly the nucleus accumbens, NAcc) also playing a key role in reinforcement learning, in particular predicting, anticipating, and estimating positive reinforcer value and any reward errors (Knutson *et al.*, 2001) in response to a variety of stimuli including drugs (Breiter *et al.*, 1997; Breiter & Rosen, 1999; Robinson & Berridge, 2003; O'Doherty, 2004) as well as the more abstract positive reinforcers such as money and humour (Knutson *et al.*, 2001; Mobbs *et al.*, 2003; Pessiglione *et al.*, 2006; Roiser *et al.*, 2009). Activity occurs in conjunction with the basal forebrain, VTA, thalamus, insula, cingulate, hippocampus and amygdala, the later demonstrated to encode stimulus reinforcement value, including the valence (Morrison & Salzman, 2010), magnitude (Bermudez & Schultz, 2010), and intensity of a reinforcer (Anderson & Sobel, 2003). Given the involvement of these structures in bvFTD as discussed, and SD (in SD seeding structural

connectivity measures from the left anterior temporal pole demonstrates functional connectivity with the subgenual cingulate/ ventral striatum and amygdala (Seeley *et al.*, 2009) a priori aberrant reinforcement learning seems likely.

Indeed, although there has been little neuroanatomical correlation of behaviours suggestive of disrupted reinforcement learning in FTD so far, what there is supports this notion. For example, over-eating and the evolution of a sweet tooth have been associated with atrophy of the right anterior insula, striatum, OFC and hypothalamus (Whitwell *et al.*, 2007; Woolley *et al.*, 2007; Hornberger *et al.*, 2010b) and in a combined cohort of bvFTD subjects with altered eating, drug or alcohol use or hypersexual behaviours, correlation with atrophy in the right ventral putamen and pallidum has also been demonstrated (Perry *et al.*, 2014). Whilst the symptoms of altered food preference, substance misuse and hypersexuality correlate with atrophy of key regions in positive reinforcement processing circuitry, and demonstrate that symptoms of FTD indicate specific problems with reinforcement learning rather than simply reflecting generalised behavioural deficits, there is another key stimulus yet to be explored, a stimulus that carries huge pleasure, despite no obvious biological purpose; namely music. A growing body of evidence from imaging in healthy subjects and the study of patients with focal brain damage suggests that the pleasure one gains from listening to music is underpinned by the same brain reinforcement learning systems as those implicated in reinforcement learning from food, sex and drugs (Breiter *et al.*, 1997; Blood *et al.*, 1999; Blood & Zatorre, 2001). Deficits in music emotion recognition and attribution of theory of mind to musical pieces have been shown in both bvFTD and SD (Hsieh *et al.*, 2011; Omar *et al.*, 2011a; Hsieh *et al.*, 2012a; Downey *et al.*, 2013). However, deficits in musical pleasure processing have yet to be demonstrated. If symptoms of FTD reflect abnormal reinforcement learning, it seems likely that abnormal responses towards music should occur and anecdotally from the bedside there are behaviours in FTD populations, such as musicophilia, suggestive of this such that assessment of music liking may provide an avenue for measuring reinforcement learning deficits in FTD (Fletcher *et al.*,

2013).

1.5.3 **Impairment of central control of autonomic effector mechanisms**

There has been a wealth of information from both animal and human studies investigating the normal central control of autonomic function. Overall interpretation is complicated, however, by the variety of techniques used for eliciting responses (isometric forced expiration, isometric hand grip, mental arithmetic, pharmacological alpha receptor stimulation to name a few) and the methods of measuring changes in autonomic arousal (variability in heart rate (HR), blood pressure (BP), galvanic skin conductance responses (SCR)). However, converging data implicates a few key anatomical areas in autonomic response generation, irrespective of the response type. Baseline fluctuations in SCR are associated with functionl magnetic resonance imaging (fMRI) changes in areas including the orbitofrontal cortex (OFC) and anterior insula (Critchley *et al.*, 2000b), and the magnitude of SCR changes elicited by sudden highly salient loud tones in healthy adults correlates with activity in areas including the anterior insula, ACC and TPJ (Mueller-Pfeiffer *et al.*, 2014), Changes in similar brain regions are observed with increases in blood pressure stimulated by isometric exercise from hand-grip or Valsalva, mental arithmetic, or pharmacological alpha-receptor stimulation with additional activation in the ACC, bilateral insular and prefrontal cortex (King *et al.*, 1999; Critchley *et al.*, 2000a). A recent meta-analysis of 43 studies of central autonomic processing in healthy individuals demonstrated that the areas most consistently activated (irrespective of method of stimulation or measurement of response) included the amygdala, right anterior insula, left posterior insula and cingulate cortex (Beissner *et al.*, 2013).

Studies in disease have revealed that medial temporal lobe resection can result in uncoupling of perceptual and autonomic reactions to stimuli. A lateralisation effect has also been suggested with patients with right medial temporal lobe resection demonstrating

diminished SCR to emotive visual stimuli whilst psychological interpretations are maintained, with the converse pattern demonstrated with left medial lobe resections (Glascher & Adolphs, 2003). In addition, patients with degeneration of the autonomic nervous system secondary to primary autonomic failure, who cannot mount normal physiological responses to emotive stimuli, have decreased amygdala activation on fMRI (Critchley *et al.*, 2002).

There has been relatively little work investigating the effects of dementia on the autonomic system and comparison of results on syndromic levels have so far been confounded by the combining of bvFTD and SD subgroups into a general 'FTLD' group in some studies (Sturm *et al.*, 2006; Werner *et al.*, 2007; Hoefer *et al.*, 2008) or use of difference metrics for measuring responses; some authors have evaluated SCR alone (Joshi *et al.*, 2014) whilst others have created 'autonomic scores' composite from SCR, HR and BP (Sturm *et al.*, 2006; Werner *et al.*, 2007; Sturm *et al.*, 2008; Sturm *et al.*, 2011). However, given the key involvement of areas such as OFC, Insula, ACC and amygdala in FTD and AD, derangements of autonomic processing are to be expected, and one may hypothesise that different syndromes will carry different signatures. For example, one could predict that patients with more dorsal disease (for example PNFA), with the least anterior-mesial or fronto-insula damage, would display the fewest abnormalities of autonomic reactivity.

1.6 **Unresolved problems:**

1.6.1 **Current measurement techniques are insensitive and/or non-specific:**

Current measurement techniques in FTD remain limited in their sensitivity to detect disease onset and progression and their ability to map underlying pathology. For example, whilst measuring cerebrospinal fluid (CSF) levels of proteins AB-142 and tau are useful in the diagnosis of AD, there are currently no useful serum biomarkers for FTD pathology (Kasai *et al.*,

2009). From a neuroimaging perspective, although individual clinical syndromes have been shown in association with characteristic atrophy patterns, such patterns are usually variable and unreliable on an individual level, a fact reflected by the supportive rather than essential role of abnormal imaging in the diagnosis of FTD (Rascovsky *et al.*, 2007; Rascovsky *et al.*, 2011). This heterogeneity is perhaps best illustrated with the bvFTD phenotypic spectrum where so-called 'phenocopy' cases exist; individuals who despite a highly suggestive clinical history have normal neuroimaging (Hornberger *et al.*, 2009; Hornberger *et al.*, 2010a; Kipps *et al.*, 2010). Although some cases may be non-neurodegenerative in origin, this phenotype has been demonstrated in bvFTD secondary to known underlying genetic (C9orf72) abnormalities (Khan *et al.*, 2012). Neuroimaging signatures of specific underlying genetic mutations including MAPT, GRN and C9orf72 are reported, although again, these are variable and often non-specific (Beck *et al.*, 2008; Chan *et al.*, 2009; Rohrer *et al.*, 2010; Becker *et al.*, 2012; Mahoney *et al.*, 2012a; Whitwell *et al.*, 2012). Characteristic patterns of changes in white matter fibre tract integrity using diffusor tensor imaging (DTI) are also reported for different clinical syndromes or genetic abnormality with some changes on longitudinal analysis but numbers necessary to demonstrate change are high (Borroni *et al.*, 2007; Matsuo *et al.*, 2008; Ash *et al.*, 2010; Krueger *et al.*, 2010; Acosta-Cabronero *et al.*, 2011; Galantucci *et al.*, 2011; Josephs *et al.*, 2011; Agosta *et al.*, 2013; Mahoney *et al.*, 2014).

1.6.2 **Optimal stimuli remain elusive:**

Generating sensory stimuli with which to best to elicit measureable responses remains problematic. Emotional responses have been induced with the use of simple startle tones, emotive music, film clips or even making experimental participants watch themselves in embarrassing situations (Sturm *et al.*, 2006; Werner *et al.*, 2007; Sturm *et al.*, 2008; Omar *et al.*, 2010; Hsieh *et al.*, 2011; Omar *et al.*, 2011a). However, the use of non-verbal auditory

information as a sensory probe is perhaps particularly germane to language led disorders, where sound perception deficits have been demonstrated to occur at different levels of an auditory processing; a perceptual level in PNFA, an apperceptive level (that is the ability to identify an auditory object when presented from a non canonical 'view point', analogous to the Visual Object Decision Task (Warrington & James, 1991)) in both PNFA and SD and a generalized semantic level in SD (Goll *et al.*, 2010).

1.6.3 **Physiological responses are difficult to assess:**

Responses towards emotional stimuli have been assessed in a variety of ways including recording of changes in facial expression, however objective measures have rarely been used and measuring dynamic changes in autonomic reactions towards stimuli may provide one such avenue for further investigation.

Decreased baseline autonomic reactivity has been demonstrated in bvFTD (Joshi *et al.*, 2014; Robles Bayon *et al.*, 2014; Struhal *et al.*, 2014) and AD (Femminella *et al.*, 2014; Struhal *et al.*, 2014), with the latter occurring early in the clinical disease courses, or even at pre-symptomatic stages (Collins *et al.*, 2012), which may suggest disruption of autonomic effector pathways. Alterations in autonomic reactions towards startling noises (single loud auditory tones) have also been examined (again using combined patient groups) with SCR demonstrated to be depressed in one study (Hoefer *et al.*, 2008) but retained in another (Sturm *et al.*, 2006; Joshi *et al.*, 2014); the reasons for these discrepancies remaining unclear. Startle responses are largely brainstem mediated (Saper, 2002), and it has been suggested that relative preservation of startle responses may reflect integrity of these structures in early disease (Seeley, 2010). However, in fMRI work in young healthy controls, the magnitude of skin conductance responses to startle has been shown to correlate with activity in the anterior insula and ACC (Mueller-

Pfeiffer *et al.*, 2014) rather than brainstem, and one may therefore anticipate impaired effector mechanisms in bvFTD and predict depressed responses. Decreased autonomic responses have also been demonstrated in bvFTD and SD in response to the more emotionally complex salience of embarrassment (watching oneself singing karaoke) (Sturm *et al.*, 2006) and in SD in response to emotionally charged conversations (discussing relationship difficulties) (Sturm *et al.*, 2011) but have conversely also been shown to be normal whilst watching emotionally charged film extracts (Werner *et al.*, 2007). Therefore there is some evidence of abnormal physiological reactivity in FTD but it is mixed and interpretation is problematic. Only one study so far has investigated the neuroanatomical correlates of depressed startle responses showing correlation with atrophy in the amygdala, ACC, OFC and Insula (Hoefer *et al.*, 2008). Therefore, autonomic reactivity profiles may differ between FTD groups, with deficits arising either in information processing or effector autonomic pathways. Potentially these could uncouple and measurement of these may offer a way to differentiate syndromes. Measurements of autonomic reactivity potentially offer an objective measure of salience responses that does not depend upon cognitive tasks and may be particularly appropriate in cognitively impaired patient groups.

Throughout this thesis I have used pupillometry as the metric of autonomic responses. Pupilliometry offers certain advantages over other correlated electrodermal and cardiorespiratory indices of sympathetic response (Granholm & Steinhauer, 2004; Bradley *et al.*, 2008; Steiner & Barry, 2011): pupil dilatation responses are relatively resistant to disease-associated movement and other artefacts, well preserved to auditory stimuli in healthy older individuals (Zekveld *et al.*, 2011) and track neural responses closely (Siegle *et al.*, 2003; Gray *et al.*, 2009; Murphy *et al.*, 2013). Moreover pupillary changes are themselves a potent reciprocal source of emotional and social signals (Harrison *et al.*, 2006; Harrison *et al.*, 2009) and the brain effector mechanisms are likely to be intimately related to core neural circuitry mediating integrated physiological responses to sensory stimuli (Beissner *et al.*, 2013; Dutta & Gutfreund, 2014; Netser *et al.*, 2014). Finally, the time course of pupillary reactions are of the magnitude of

a few seconds rather than tens of seconds for other autonomic metrics allowing for shorter experiments to make working with behaviourally challenged subjects more feasible.

1.7 Structure of this Thesis:

Outlined above is evidence to suggest that many of the symptoms observed FTD may reflect abnormalities in physiological processing systems. These may break down on three main levels; firstly, coding of afferent sensory information, secondly, assignment of appropriate salience and pleasure to allow interpretation of this information in an appropriate emotional context, and thirdly, generation of effector autonomic responses. As different FTD syndromes bear the brunt of disease in different anatomical regions, different systems may be differentially affected and may display syndrome specific deficits in normal autonomic reactivity to salient information. Investigation of the anatomical underpinning of little studied symptoms that may speak to deranged sensory processing will provide direction for future investigation and by probing FTD syndromes using a range of auditory experimental tools designed to manipulate the salience and affective parameters, different physiological signals may be produced for each group, which can then act as in-vivo dynamic biomarkers.

Outlined below are five experiments designed to probe the different levels of these processing pathways: the first investigates abnormal sensory coding; the second, third and fourth examine the use of physiological metrics to explore three levels of abnormal salience processing, that derived from low-level auditory cues, more complex emotional salience and finally the unexplored potential salience of semantics; the final experiment investigates abnormal music processing as a model of disrupted reinforcement learning systems.

Experiments:

1.7.1 Experiment 1: *Do symptoms suggestive of altered sensory perceptions in FTD and AD reflect abnormalities of core sensory coding pathways?*

Behaviours suggestive of abnormalities of pain and temperature perception are anecdotally common in FTD but have received little attention in the literature and it remains to

be established whether any abnormality observed is part of a generalized disordered behavioural syndrome, or speaks to more specific derangement of physiological sensory signal coding. In this experiment information obtained from the carers of FTD patients was used to better characterize the nature of these symptoms in comparison and contrast to patients with AD. Using voxel-based morphometry (VBM) analysis, neuroanatomical correlates of abnormalities in pain and temperature perceptions were then identified.

Here I hypothesised that patients with FTD syndromes will demonstrate deficits in sensory (pain and temperature) processing that would reflect focal atrophy to specific brain regions involved in normal processing of these sensory perceptions. I predicted that in the patient groups, SD and bvFTD, with a greater disease burden to the fronto-insular region, these symptoms would occur more frequently. Further, I predicted that abnormal symptoms will reflect damage to specific brain regions involved in the generation of normal pain and temperature perceptions.

1.7.2 Experiment 2: *Is salience encoding from primitive cues disrupted in FTD and AD?*

Salience information from a stimulus is potentially carried by an array of low-level sensory cues such as contrast and proximity. In this experiment the effects of disease upon differential effects of perceived motion direction as measured by pupillary reactivity was examined. Since previous work has implicated the amygdala as a key area in processing of the greater salience effect of approaching relative to withdrawing sounds as a 'warning cue' heralding danger, I hypothesised that SD, showing greater amygdala damage relative to the other syndromes, would display loss of this normal effect and that this could be used to physiologically to differentiate SD from other FTD syndromic groups.

1.7.3 Experiment 3: *A disruption of salience processing from more complex emotional cues in FTD and AD?*

Emotional processing deficits are common to neurodegenerative diseases and may speak to core impairment in the ability to extract or assign salience to usually emotionally stimuli.

This was investigated here by measuring pupillary responses towards a battery of emotionally charged non-verbal sounds. I hypothesised that with greatest damage to fronto-insular regions, areas key to autonomic reactions, bvFTD would show overall depressed autonomic responses to auditory stimuli, irrespective of valence. I further predicted that the syndromes of bvFTD and SD, showing greatest emotional recognition deficits, would display the greatest impairment in physiological reactivity as modulated by emotional valence and that a normal relationship between pupillary response and valence of sound would be preserved in both PNFA and AD. Further, I predicted that bvFTD, having the greatest damage to the fronto-insular region, would demonstrate the most deranged physiological reactions and that in SD deranged responses would potentially reflect a damaged semantic system rather than impaired core autonomic reactivity.

1.7.4 Experiment 4: *A disruption of salience processing from semantic evaluation in FTD and AD?*

The ability to disambiguate perceptually similar auditory objects in the environment and assign attentional resources appropriately likely relies upon the semantic system, and disruption of this system may lead to inappropriate salience assignment. There has been little work so far investigating whether salient information is carried by the semantic content of a stimulus and how this may be disrupted in health and disease.

In this experiment, physiological responses and neuroanatomical correlates of differential responses to meaningful and perceptually matched meaningless sounds were investigated.

I hypothesised that usually meaningful sounds would evoke greater pupillary responses than acoustically matched meaningless counterparts and that patients with impaired semantic function would demonstrate a loss of this normal differential response. This would provide a physiological metric of semantic ability with the potential to differentiate and track disease progression.

1.7.5 Experiment *5: Are abnormal pleasure responses to environmental sounds and music underpinned by disrupted reinforcement learning in FTD and AD?*

Disrupted behaviours observed in FTD and AD in response to sounds, both environmental and music, are under recognised and may suggest that patients have distorted processing of the normal pleasantness of sounds. I hypothesised that such behaviours are actually relatively common, can dissociate by sound type, and are underpinned by damage to brain regions involved in normal reinforcement learning such that emotional salience processing is disrupted. Here, using VBM, the neuroanatomical basis of behaviours suggestive of abnormal hedonic processing of sounds and music was addressed in a series of patients with FTD and AD. Quantitatively the regional brain atrophy patterns of those who did with those who did not exhibit abnormal behaviours were compared to investigate the anatomical substrates of altered sound reinforcement learning.

Chapter 2. General methods and cohort characteristics

2.1 Patient details:

2.1.1 Background demographics:

A total of 160 subjects participated in the work included in this thesis. Background neuropsychometric scores and details of study participation for each individual can be found in table 10.2 in Appendix. The details of the baseline demographics for each study group are given in the relevant experimental chapter. All patients included met current criteria for diagnosis (Dubois *et al.*, 2007; Rascovsky *et al.*, 2007; Gorno-Tempini *et al.*, 2011) based upon clinical syndrome as determined by a senior neurologist. Healthy older individuals who participated as controls had no history of neurological or psychiatric illness. All participants were recruited consecutively via the specialist Cognitive Disorders Clinic at the National Hospital for Neurology and Neurosurgery. Recruitment and testing occurred over a three-year period. No participant had a clinical history of hearing loss or pupillary disease and none was considered to have clinical evidence of a mood disorder at the time of participation. In all experiments, the groups were matched for age, disease duration and level of educational attainment. 43 patients had undergone genetic testing (21 bvFTD, 12 SD, 9 PNFA) and this revealed an underlying genetic mutation in 14 cases (8 C9orf72, 6 MAPT, all of whom had presented with bvFTD, apart from one C9orf72 case who had a clinical syndrome of PNFA). Cerebrospinal fluid tau and beta-amyloid assays (7 bvFTD, 4 SD, 6 PNFA, 18 AD) and Florbetapir positron emission tomography (PET) brain imaging (3 PNFA, 5 SD) (where available) further corroborated the clinical diagnoses (CSF total tau: beta-amyloid ratio >1 in all AD cases, Florbetapir-PET negative for amyloid deposition in available SD and PNFA cases). 6 patients were taking anti-depressant (1

AD, 2 PNFA, 2 SD, 1 bvFTD) and 17 were taking anti-cholinesterase inhibitor medication (9 AD, 2 PNFA, 1 SD, 5 bvFTD).

All experimental work in this thesis was approved by the UCL Institute of Neurology and National Hospital for Neurology and Neurosurgery Joint Research Ethics Committee and written informed consent was obtained for all participants in accordance with the Declaration of Helsinki.

2.1.2 Background neuropsychological evaluations:

Participants underwent a battery of baseline neuropsychological tests prior to participation in the experiments. General Verbal and Performance IQ were estimated using the Wechsler Abbreviated Scale of Intelligence (WASI; Wechsler, 1999), which is an abbreviated version of the Wechsler Adult Intelligence Scale. Episodic memory was estimated using The Recognition Memory Test for faces/words (RMT; Warrington, 1984). In this test 50 pictures of faces or printed words are presented. After a delay the participant is presented with a target pair of words or faces, one of which will have been previously seen. In a two-alternative forced choice paradigm the participant is required to select the previously presented stimulus. Naming was assessed using the Graded Naming Test (GNT; McKenna & Warrington, 1983) - which requires participants to name line drawings that become of increasingly lower frequency. Semantic memory was assessed in patients using The British Picture Vocabulary Scale (BPVS; Dunn, Dunn & Whetton, 1982) – in which participants are presented with a written word (which was also spoken) and four pictures, one of which corresponds to the word and must be chosen. This is of graded difficulty and has a normal ceiling effect in healthy adults. This tests avoids confounds of lexical retrieval naming deficits. In addition, verbal semantic comprehension was assessed more specifically with an in house devised synonym matching task (where a participant is presented with three written words and must select which of the second or third word aligns most closely with the first (Warrington, McKenna, Orpwood., 1998). Visuo-spatial perceptual function was assessed with the use of The Object Decision subtest of

the Visual Object and Space Perception battery (VOSP) (Warrington & James, 1991) – in which participants are required to identify a 75 degree rotated real object from three nonsense silhouettes of similar complexity. Working memory was assessed using Digit Span tasks (Wechsler, 1987) where number strings of increasing length must be repeated (forward condition) and then had to repeat the numbers in the opposite order to that in which they were presented by the examiner (reverse condition) as a marker of executive function. To avoid linguistic confounds, executive function was also estimated with a measure of spatial span backwards in which the participant must point to 3D blocks on a board in the reverse order to which they were presented (the CORSI test). Additionally, pre-potent response inhibition as a further test of executive function was assessed on The D-KEFS Colour Word Interference Test (DKEFS Stroop; Delis, Kaplan & Kramer, 2001) - which is an adaptation of the Stroop test, where colour words are presented in a conflicting ink colour and participants are timed naming the colour of the ink for 50 of these words.

2.2 Experimental design and methods:

2.2.1 Assessment of auditory semantic function:

In order to measure baseline auditory semantic function, independent of naming or cross-modal labelling, I devised a nonverbal sound semantic classification task adapted from a previously described procedure (Goll *et al.*, 2010). Nonverbal sounds were derived from publically available sound libraries (www.freesound.org, www.freesfx.co.uk) and sampled a range of human, animal, environmental and mechanical sounds. In a pilot experiment, 20 healthy younger individuals (median age 28 years (range 23-37), six male) were asked to identify an initial set of 180 sounds: those that were misidentified by two or more (>10%) of the healthy younger pilot control group were excluded in order to ensure all sounds in the main experiment were intrinsically highly familiar and identifiable. Young healthy controls were initially used as these were easy to recruit from within the research department. Audio samples

were converted to digital wavefiles, and edited in Goldwave© so that all environmental sound stimuli were five seconds in duration (brief sounds such as hiccoughs that tend to be naturally periodic were repeated within this interval). Using a program previously written in Matlab© mean overall intensity (root mean square value) was fixed across stimuli to ensure that all stimuli were of similar loudness. Sounds were combined such that the constituent sounds were associated either with the same sound source (e.g. a goose honking, a goose's wings flapping) or with different sources (e.g., a goose honking, a child yawning). Sound pairs in the 'same' and 'different' conditions did not differ systematically in the acoustic similarity of their component sounds; sound pair classification therefore relied on a semantic decision based on recognition of the sounds and could not be based on perceptual criteria. A total of 80 sound pairs were created initially. A program was written in Matlab© to present sounds in a pseudo-randomised order; the sounds comprising each pair were presented serially with a 1 second inter-sound delay. The task on each trial was to decide whether the paired sounds came from the same source or different sources ('Are the sounds made by the same thing or different things?'), and the participant could respond verbally or by pointing to the corresponding written word on a prompt sheet. All data was recorded in Matlab for off-line analysis. No feedback about performance was given and no time limits on responses were imposed. Sound pairs incorrectly labelled by more than 10% of the healthy older controls were deemed to be inherently too ambiguous and were excluded. This yielded a total of 60 sound pairs used in the final stimulus set shown in table 2.2.

2.2.2 Assessment of peripheral hearing function:

Peripheral hearing ability was assessed using pure tone audiometry. On a separate occasion to the main experimental testing, participants were seated in a quiet room and pure tones were played via headphones from a laptop computer. Subjects were permitted to keep any hearing aids in place throughout the testing (and main experiment). The procedure was

adapted from a commercial screening audiometry software package (AUDIO-CDTM®, http://www.digital-recordings.com/audiocd/audio.html). Five frequency levels (500, 1000, 2000, 3000, 4000 Hz) were assessed: at each frequency, participants were presented with a continuous tone that slowly and linearly increased in intensity. Participants were instructed to indicate as soon as they were sure they could detect the tone and response times was measured and stored for offline analysis. Hearing was assessed in each ear in each participant and a mean measure of hearing (averaged from thresholds at the different frequencies and between ears).

'different' sound pairs		'same' sound pairs	
baby cooing	stream burbling	baby cooing	baby laughing
baby laughing	waves lapping	car crash sounds	car horn
car alarm disarmed	shovel on metal	car horn	car skidding
car horns	grandfather clock ticking	child yawning	child hiccupping
car skidding	man snoring	clock ticking	clock alarm bell
cat whining	puppy yelping	cockerel crowing	hen clucking
child yawning	geese honking	geese honking	goose wings flapping
clock ticking	baby crying	grandfather clock chime	grandfather clock ticking
cuckoo clock	man breaking wind	horse whinny	horse hooves
dog growling	female cough	Infant wailing	Infant sobbing
dog lapping	hiccupping	man clearing throat	man sighing
fizzy drink can opened	coin dropped on table	man shouting in pain	man vomiting
hen clucking	man sighing	man sobbing	person breaking wind
horse trotting	person walking on gravel	person coughing	person snoring
horse whinnying	woman yawning	pigeon wings flapping	pigeon cooing
infant sneezing	engine running	stream burbling	waves lapping
infant wailing	man wheezing	telephone ringing	dialling tone
man shouting in pain	car horn	train travelling on tracks	train horn
man sobbing	bees humming	trickling water	crashing waves
man vomiting	clock alarm bell	woman clearing throat	woman yawning
mosquito	woman screaming	woman crying	woman screaming
paper rustling	woman clearing throat	woman giggling	woman humming
paper tearing	child hiccupping		
person brushing teeth	train horn		
person clapping hands	dog barking		
phone engaged tone	cat hissing		
pigeon cooing	Fingers clicking		
pigeon wings flapping	person chewing		
shovel digging gravel	car window winder		
small bird flapping wings	graveyard Wind		
telephone dial tone	man clearing throat		
telephone receiver replace	female coughing		
telephone ringing	person breathing		
thunder	car engine starting		
train travelling on tracks	baby cough		
waves crashing	woman giggling		
woman crying	car crash sounds		
woman humming	water trickling		

Table 2.1. Sound pairs used in the semantic matching test: Sounds in each pair were presented at fixed mean intensity and serially with a 1 second inter-sound gap; sound pairs were presented in randomised order. The task on each trial was to decide if the source of each sound was the same or different ('Are the sounds made by the same kind of thing or different kinds of things?")

2.2.3 Pupillometry experiments:

2.2.3.1 Design:

For pupillometry measurements trials were generally designed such that pupil recording would occur for an initial silent interval (two seconds), prior to sound stimulus onset. From this pupil areas could be averaged to produce a baseline from which to measure subsequent change. This was then followed by the sound stimulus (five seconds for experiment 5 and two seconds for experiments 3 and 4) plus a further silent period of pupil recording (7 seconds) in which the pupil response could equilibrate (the optimal length of this equilibrium period was estimated to be seven seconds from examining traces in pilot recordings in young healthy controls and was aimed as a balance between collecting insufficient data versus experiments becoming too long for tolerability in patients with behavioural issues). Following termination of recording, a modified Likert scale was displayed in which the participant would, with no time constraints, rate the sound that they had just heard for affective valence 'how pleasant was the sound' and then for alerting rating 'how alerting did you find the sound?' using a wireless mouse. Trial design is schematised in Figure 2.1.

2.2.2.2 Pupillometry data acquisition:

For all pupillometry experiments, sound stimuli were presented via high-fidelity headphones (ATH-M50 Audio-Technica®) from a notebook computer at a constant, comfortable listening level (at least 70 dB). During pupillometry, participants were seated before a computer monitor in a dimly but uniformly illuminated room.

Pupil area was measured from the right pupil using an infra-red camera (Eyelink II; SR Research, Canada) mounted on a headset just below the line of sight while the participant fixated a 1 cm white circle in the centre of the monitor screen. All experiments were run using Eyelink II software. Each experimental trial was triggered once adequate visual fixation was

achieved and pupil area was measured (sampling rate 250 Hz) over the entire trial duration. All pupil response and behavioural rating data were recorded by the eyelink© software for off-line analysis.

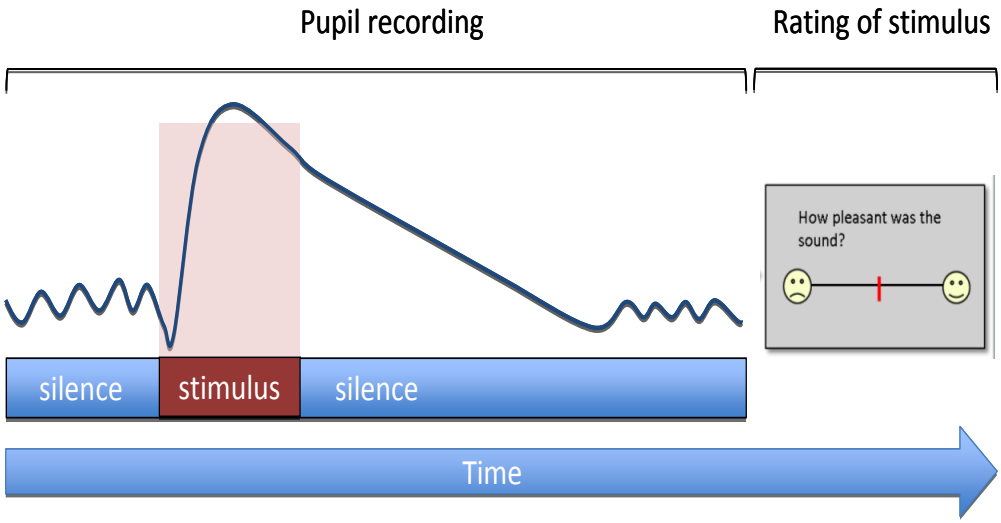

Figure 2.1. Schematic of experimental trial design. Pupillary sizes are recorded for two seconds of silence, followed by stimulus presentation (shaded box) for five seconds and then a further 7 seconds of silence where pupil responses continue to be recorded before time unrestrained ratings on a likert scale.

2.2.2.3 Data conditioning

Data were transferred from the Eyelink© desktop computer as excel files and initially imported into STATA 12.1 for processing. A program was written to perform the following steps prior to analysis of pupil responses:

2.2.2.3.1 ***Removal of artifacts secondary to blinking:***

Loss of pupillary recordings secondary to blinking is a common potential source of artefact in experiments. Any complete data losses (these are marked by 0 in the eyelink© software) were identified and extracted including 25ms prior to the data loss.

As a blink is followed by a normal light reflex in which there is an initial pupil dilation following eye closure and then a compensatory pupil constriction, failure to extract these responses adequately could lead to overestimation of pupil area. In order to determine the optimal amount of data removal in order to ensure complete removal of the affected area without excessing loss of data, data was collected in 10 healthy young controls whilst they voluntarily blinked for varying lengths of time. The length of the light reflex did not appear to correlate with blink length and I determined that removal of data for 750 m/s following a blink would capture all light reflex reactions without excessive data extraction, irrespective of the length of the blink.

2.2.2.3.2 ***Identification and extraction of anomalous data regions:***

in addition to areas of complete data loss secondary to blinking, examination of traces of pupil size over time revealed areas of data loss secondary to partial obscuration of the pupil occurring when subjects had partially closed their eye or looked away from the camera to such a degree that that an accurate recording of pupil area could not be obtained. Unlike blinks, which had a characteristic waveform and could easily be recognised by the software, these areas of data loss were more heterogeneous in form and more problematic to identify. An algorithm was designed to recognise the start of a period of data loss by identifying a rapid decrease in pupillary area (-15 units relative to the data point prior over two data points) if the pupil area was relatively stable prior to this (two data points prior to this 'start' were fluctuating by less than 10 units). This determination of 15 units change per data point as the optimal sensitivity to extract an obscuration without excessive data loss was determined manually by examining traces of pupil size over time after attempting extractions of different thresholds. The 'end' of an anomalous region was identified if a region (following the 'start' of data extraction) increased by more than 8 units successively over two data points and then levelled off. No change less than 10 units in total length was extracted.

2.2.2.3.3 ***Calculation of pupil response:***

Previous work has usually taken the change in pupil dilatation from baseline as the measure of response to a given stimulus. In early experiments, prior to the advent of computing, the pupil was simply photographed at regular intervals following onset of a stimulus and pupillary areas manually traced and plotted over time (Hess & Polt, 1960; 1964; Kahneman & Beatty, 1966) and in more recent works, algorithms have been developed to extract areas of data loss secondary to blinking prior to calculation of pupillary changes. From reading the literature it remained unclear to me at the start of this project, however, whether this methodology was used because it was optimal or because it had followed a natural evolution from these important earlier works. One could hypothesise that there would be potentially further information to be gleaned from investigating (a) the velocity of the initial upstroke of the pupillary dilatation and (b) the duration of the response in addition to the maximal pupillary dilation; more physiologically evocative stimuli could potentially evoke faster and more sustained responses as well as responses of greater overall magnitude. Therefore, in the initial iteration of the program, we calculated the 'area under the curve' as a response measure. Following extraction of blinks and anomalous regions, missing data points were interpolated. A 5 point band-pass filter was used to smooth the data and all changes in pupil size over time from a baseline were integrated. However, after these measures, examination of pupil size/time traces still revealed error in the extraction of anomalous regions areas of data that had not been extracted fully with the above methods. Extraction of excessive areas of data loss made the final calculation of the area under the curve unreliable. Extraction of insufficient data resulted in under-estimation of the area under the curve. Theoretically, manual extraction and interpolation of missing data could have been performed but this was unviable for two main reasons; firstly, in the three pupillary experiments discussed in this work, there was a total of over 4,200 trials collected, making this unfeasible, secondly, in some cases the form of

the raw trace was so degraded by artefact that the result would have been in many cases estimation only and highly subject to individual bias. Whilst the area under the curve method was highly vulnerable to underestimation of pupillary area secondary to incomplete extraction of obscuration artifacts, measurements of maximal pupil size was less vulnerable and was used in the final analysis. To limit the cumulative effect of artifacts, data was truncated to five seconds from sound onset as all pupil maximal responses occurred within the first two seconds.

2.2.2.3.4 ***Determination of baseline:***

Baseline values were calculated as the mean pupil area between 200 m/s and 2.2 seconds of trial recording (the latter point was where the initial dilation of the pupil in response to the sound stimulus occurred).

2.2.2.3.5 ***Allowance for baseline area:***

Maximum pupil area during a given trial was positively correlated with baseline pupil area. In order to avoid this potentially confounding influence, the log ratio of maximal pupil area to baseline pupil area was used as the metric of pupil response for each trial (Pupilmax).

2.2.3 **Analysis of behavioural and physiological data:**

All data were analysed in STATA 12.1®. In all analyses, a threshold $p<0.05$ was accepted as the criterion for statistical significance. To compare group demographic data, chi^2 tests were used for categorical data and linear regression for continuous variables. A one-way ANOVA was performed to determine the presence of systemic group differences before comparing specific groups with linear regression. Before calculating mean pupil responses for analysis, individual pupil responses to each sound were adjusted for any potential confounding effects of sound loudness as this is known to drive pupillary reactions (Goldwater, 1972). As pupillary responses

could potentially be systematically affected by position of a stimulus within a play list (for example, fatigue, habituation), a linear mixed effects model with crossed random effects for participant and sound was used for analysis (xtmixed pupil-response regression-variable || _all: R.name || _all: R.trialindex if group == "x"). Where a single response was generated for each individual (score on the semantic matching task, mean group pupillary responses to a given condition, following initial one-way ANOVA to demonstrate the presence of systematic differences, linear regression was used to compare group and correlations with disease severity metrics (symptom duration and a general executive measure, reverse visual spatial span) were assessed using linear regression models.

2.2.4 **Brain image acquisition and analysis:**

Sagittal 3-D magnetization-prepared rapid-gradient-echo T1-weighted volumetric brain MR sequence (echo time/repetition time/inversion time ¼ 2.9/2200/900ms, dimensions 256x256x208, voxel size 1.1x1.1x1.1 mm) was acquired on a Siemens Trio 3 tesler MRI scanner using a 32-channel phased-array head-coil. Pre-processing of brain images was performed using the New Segment (Weiskopf *et al.*, 2011) and DARTEL (Ashburner, 2007) toolboxes of SPM8 (www.fil.ion.ucl.ac. uk/spm) under Matlab 7.0® and following an optimised protocol (Ridgway *et al.*, 2008). Normalisation, segmentation and modulation of grey and white matter images were performed using default parameter settings and grey matter image were smoothed using a 6mm full width-at-half-maximum Gaussian kernel. Study-specific template mean brain images were created by warping all bias-corrected native space brain images to the final DARTEL template and calculating the average of the warped brain images. In order to adjust for individual differences in global grey matter volumes during subsequent analysis, total intracranial volume was calculated for each patient by summing grey matter, white matter and cerebrospinal fluid volumes following segmentation of all three tissue classes. Separate voxel-wise linear regression models were used to assess associations in the patient cohorts between regional grey matter volume and parameters of interest for each study. Age, total intracranial

volume, disease duration, and syndromic group membership were included as covariates of no interest in each model for all studies. To help protect against voxel drop-out because of potentially marked local regional atrophy in particular scans, I applied a customised explicit brain mask based on a specified 'consensus' voxel threshold intensity criterion (Ridgway *et al.*, 2009), whereby a voxel was included in the analysis if grey matter intensity at that voxel was > 0.1 in >70% of participants (rather than in all participants, as with the default SPM8 mask). Statistical parametric maps of regional grey matter volume correlating with behavioural response parameters of interest were examined at threshold $p<0.05$ after family-wise error correction for multiple voxel-wise comparisons within regional volumes of interest, based on my prior anatomical hypotheses. Anatomical small volumes were created by manually tracing from the template mean brain image using MRICron® (http://www.sph.sc.edu/comd/rorder/mricron.html). MRICron® is a cross-platform image viewer. This program allows the importation, viewing, rendering, and drawing of anatomical regions of interest on brain images. MRICron was used within this thesis to import and draw anatomical regions of interest to be used as volumes of interest to mask data and restrict statistical analyses to pre-defined areas of interest within VBM analyses. Anatomical regions were customised from the Oxford/Harvard brain maps in FSLview v3.1 (Desikan *et al.*, 2006; Jenkinson *et al.*, 2012) to fit the group mean template brain image in each study. All images in this thesis are displayed on sections of a group mean T1-weighted MR brain template image in MNI standard space; the left hemisphere is shown on the left of the axial section and are thresholded at $p<0.001$ uncorrected for multiple voxel-wise comparisons over the whole brain.

2.3 Background Results:

2.3.1 Auditory semantic function:

Results of the auditory semantic function test are shown for each participant in the background demographics table and group results are summarised in table 2.2.

group	Control n=25	bvFTD n=15	SD n=12	PNFA n=10	AD n=11	ANOVA
SMT score (%) (range)	90.1 (75-100; SE: 0.03)	76.8 (61.7-98.3; SE: 0.03) a	72.8 (55-85; SE: 0.03) a,b,c	83.5 (68.3-95; SE:0.03) a	82.1 (68.3-91.7; SE:0.03) a	p<0.0001, r^2=0.42, F=10.52

Table 2.2: mean group performance on the non-verbal auditory semantic matching task (SMT) with ranges in parenthesis. Initial one-way ANOVA prior to specific group comparisons is displayed in the right hand column. Performance significantly worse relative to controls, a; PNFA b; AD, c. Ranges and standard errors are in parenthesis.

All patient groups performed significantly worse than controls (controls vs. bvFTD, t= -5.10, p<0.0001; controls vs. SD t=-5.40, p<0.0001; controls vs. PNFA, t= -1.71, p=0.04, controls vs. AD, t=-1.92, p= 0.01) with the SD group significantly impaired relative to the PNFA (t= 3.24, p<0.01) and AD group (t=2.68 p=0.01). SMT scores did not correlate with standard verbal measures of semantic function (BPVS). Impaired performance in both SD and PNFA relative to healthy controls is in keeping with previous work (Goll *et al.*, 2010; Goll *et al.*, 2011) and likely reflects impairment of core semantic processing in the SD but deficits in lower level (apperceptive) sound processing in the PNFA group, in keeping with a hierarchical sound processing pathway (Griffiths & Warren, 2002; Goll *et al.*, 2010). Deficits in semantic non-verbal sound processing have not previously been demonstrated in bvFTD but were anticipated priori in light of the overlap of network involvement in bvFTD with SD.

2.3.2 **Baseline pupillary responses:**

Examining average baseline pupil responses for each participant from all three pupillary experiments, using a simple regression model, the total proportions of data points removed due to artefacts did not differ significantly between sound condition or between experimental groups. There were no effects of age, gender, medication use or disease severity upon baseline pupil size or pupillary reactivity in any group. Baseline pupil sizes were not significantly different between groups. Mean pupil response began approximately 25ms after stimulus onset. There was no correlation between baseline pupil size or reactivity and age. In all experiments, magnitude of $Pupil_{max}$ correlated with the loudness of the stimulus and this was used as a

covariate in all analysis. No systematic change in either baseline or maximal pupil change was observed with experimental progression.

Chapter 3: Pain and temperature processing

3.1 Chapter summary:

Symptoms suggesting altered processing of pain and temperature have been described in FTD and may contribute importantly to clinical phenotypes.

Better characterisation and evaluation of neuroanatomical underpinnings may shed light on specific disease processes and also provide further information on how the brain codes sensory information. Here pain and temperature symptoms were analysed using a semi-structured caregiver questionnaire recording altered behavioural responsiveness to pain or temperature for a cohort of patients with FTD and a comparison cohort of patients with amnestic AD. Neuroanatomical associations were assessed using blinded visual rating and VBM of patients' brain MR images. Certain syndromic signatures were identified: pain and temperature symptoms were particularly prevalent in behavioural variant frontotemporal dementia (71% of cases) and semantic dementia (65% of cases) and in association with C9orf72 mutations (6/6 cases), but also developed in Alzheimer's disease (45% of cases). Blunted responses were more common in the bvFTD group (40% of cases) and heightened responsiveness in the SD (73% of cases) and AD (78% of cases) groups. Within the FTD cohort, pain and temperature symptoms were particularly associated with grey matter loss in a right-lateralised network comprising posterior thalamus, posterior insula and anterior temporal cortex. This network is well established in the normal processing of sensory information in relation to homeostasis. Within the C9orf72 genetic group, pain and temperature symptoms were specifically associated with bilateral posterior thalamic atrophy. Together the findings suggest that abnormalities of sensory perceptions in FTD are underpinned by specific anatomical disruptions to sensory pathways and may reflect derangement of signal processing on a

core homeostatic physiological processing level rather than simply forming part of a generally disrupted behavioural syndrome.

3.2 Introduction:

Psychiatric symptoms including delusions, psychosis, and somatisation behaviours are increasingly recognised in FTD, especially in association with the recently discovered C9orf72 genetic mutations (Dobson-Stone *et al.*, 2012; Khan *et al.*, 2012; Snowden *et al.*, 2012; Downey *et al.*, 2013; Galimberti *et al.*, 2013). Many of these appear to reflect disruptions of the perception of the state of one's own body, or one's external boundaries (Clark *et al.*, 2014b; Downey *et al.*, 2014), and may speak to difficulties with differentiation of the external environment and internal milieu. Neuroanatomically, this may relate to thalamico-cortical circuits (Downey *et al.*, 2013) involved in gating of incoming afferent sensory information (Clark *et al.*, 2014b). In both bvFTD and SD abnormal sensory perceptions have been reported in conjunction with somatisation behaviour (Snowden *et al.*, 2001; Chan *et al.*, 2009; Landqvist Waldo *et al.*, 2014) and these too may form part of a general sensory processing problem with impairment of differentiation of the internal and external environments. Using semi-structured telephone interviews to carers of patients with both bvFTD and SD, Snowden and colleagues reported on a range of 'sensory behaviours', consisting of apparent alterations in pain and thermal awareness. Patients were categorized as demonstrating either loss of awareness of pain or exaggerated responses to both pain and thermal stimuli. More specific details were not available. These symptoms were surprisingly common occurring in around 40% of cases, with a preponderance towards a decrease in the apparent awareness of painful stimuli in the bvFTD group and exaggerated responses to both painful and thermal stimuli in the SD group and although discussed in the context of more widespread behavioural deficits, these

symptoms potentially reflect deficits on a more core physiological sensory processing level.

With a retrospective review of case notes it has also been demonstrated that somatic symptoms can occur as the presenting complaint in FTD (6 of 36 patients; 5 bvFTD, 1 PNFA) (Pijnenburg *et al.*, 2004). In this study patients reported headache, hearing noises, allergic reactions and feeling 'hypoglycemic', symptoms for which the authors could elucidate no clear underlying cause. Further, in a recent retrospective review of the case notes of 97 patients with histopathological confirmed diagnoses of FTD pathology, somatic complaints were also found to occur in around 40% of cases (Landqvist Waldo *et al.*, 2014). These were characterised by non-specific unexplained headaches (25%), musculoskeletal (14%), gastrointestinal or urogenital symptoms (12%) and abnormal pain responses in five patients (including vague migrating pains, chest pains and pruritis). Six patients showed exaggerated reactions to sensory stimuli, with allodynia in three cases such that simple tasks such as having nails or hair cut was painful, whereas other patients showed reduced response to pain (chewing on shards of glass). Hypochondriasis occurred in 16 patients, fourteen of whom also displayed somatic complaints. Unfortunately, the clinical phenotypes of these patients were not given so associations between particular behaviours and FTD sub-group phenotypic correlations could not be made.

Such reports suggest that pain and temperature responsiveness are commonly altered in FTLD and this is of considerable interest on both neurobiological and clinical grounds, potentially speaking to a more generalised disordered homeostatic sensory coding system. In the healthy brain, pain and thermal processing rely upon a network gated through the posterior thalamus and focused upon the insula cortex, with this region most consistently activated in functional imaging studies of pain processing and the only region where direct micro-stimulation in awake humans produces pain (In addition to a wide range of additional sensory phenomena) (Greenspan & Winfield, 1992; Peyron *et al.*, 2000; Craig, 2002; Olausson *et al.*, 2002; Brooks *et al.*, 2005; Hua *et al.*,

2005; Herde *et al.*, 2007; Craig, 2009; Mazzola *et al.*, 2009; Isnard *et al.*, 2011; Mazzola *et al.*, 2012; Moulton *et al.*, 2012; Meerwijk *et al.*, 2013). This network interacts with those involved in other key aspects of sensory stimuli analysis; sensory gating and representation (thalamus, somatosensory cortex, posterior insula), arousal and attention (thalamus, anterior cingulate), evaluation (anterior insula, anterior cingulate, antero-medial temporal structures), programming behavioural responses (anterior cingulate, orbitofrontal and prefrontal cortices) (Greenspan & Winfield, 1992; Peyron *et al.*, 2000; Singer *et al.*, 2004; Brooks *et al.*, 2005; Craig, 2009; Mazzola *et al.*, 2009; Isnard *et al.*, 2011; Mazzola *et al.*, 2012; Moulton *et al.*, 2012; Meerwijk *et al.*, 2013) and placing sensory stimuli in semantic and hedonic context (via an anterior temporo-orbitofrontal 'appraisal network': (Guo *et al.*, 2013; Zhou & Seeley, 2014). There have been no detailed neuro-anatomical correlations of abnormal somatosensory processing in FTD, however, given the overlap of the anatomical substrate of these networks and those targeted by the pathological processes in FTLD, deficits observed are likely to be underpinned by damage to these areas. Moreover, altered experience of pain and temperature might be also predicted to occur in AD, which targets more dorsal regions that process pain and temperature (Borsook, 2012) and regions that link the salience network and DMN (Kucyi *et al.*, 2012a). Pain and temperature alterations are not widely recognised as clinical issues in AD, and limited available information suggests that sensory encoding and perception of pain are retained, at least in early to moderate stage disease, with engagement of a similar central nociceptive network to healthy older individuals (Cole *et al.*, 2006); however, patients' pain tolerance has been variously reported as unaltered, increased or diminished (Cole *et al.*, 2006; Cole *et al.*, 2011; Borsook, 2012; Jensen-Dahm *et al.*, 2014).

Therefore if the somatic symptoms discussed in the literature are indicative of disordered physiological processing pathways, one may hypothesise that different clinical syndromes will result in selective disruption of these pathway and separable behavioural and neuroanatomical signatures within the FTD spectrum and between FTLD syndromes and AD. For example, if patients

with C9orf72 mutations have particular thalamic disease burden, and the thalamus is integral to accurate early sensory gating, the clinical signal may be different to patients with bvFTD who show maximal disease burden on the fronto-insular regions. Alternatively, those regions important for interpreting the meaning of incoming sensory information, and placing it in an appropriate semantic and emotional context (via the 'appraisal network' (Guo *et al.*, 2013)), are more affected in SD and could account for over-attribution of importance to sensory information and the exaggerated responses to painful and thermal stimuli observed above (Snowden *et al.*, 2001). One can therefore easily predict that different syndromes may have different, and potentially separable, physiological signatures of disordered somatosensory processing underpinned by overlapping anatomical substrates.

Hypotheses and predictions: Here I addressed these issues in a cohort of patients with FTLD and in a comparison cohort of patients with AD. Symptoms suggesting altered pain and temperature processing were characterised using a semi-structured proforma administered to patients' care givers. Structural neuroanatomical correlates of these symptoms were assessed using VBM patients' brain MR images. I hypothesised that patients with FTD syndromes would demonstrate deficits in sensory (pain and temperature) processing and that these symptoms would occur more commonly in those syndromes with greater fronto-insular damage, namely bvFTD and SD and in particular, in patients with C9orf72 mutations versus other disease groups.

I further hypothesised that symptoms in the FTLD cohort and in AD would be particularly associated with grey matter atrophy of the insula, as a key hub region in somatosensory coding (Peyron *et al.*, 2000; Craig, 2002; Zhou & Seeley, 2014) and that in both SD (based on previous work) and AD (on anatomical grounds) symptoms of exaggerated responses towards stimuli would be observed but that these would reflect differential involvement of the salience network.

3.3 Methods:

3.3.1 Patient characteristics:

Fifty-eight patients with a syndrome of FTD (25 female, aged 52 – 84 years) and 20 patients with AD (eight female, aged 53 – 74 years) were assessed consecutively over a three-year interval via a tertiary Cognitive Disorders Clinic. CSF and brain amyloid PET imaging corroborated clinical diagnosis (ratio of total tau: beta-amyloid$_{1-42}$ levels >1 in 14/14 AD cases and <0.8 in 13/13 FTLD cases, Flubetapir PET negative for amyloid deposition in 7/7 FTLD cases). All patients had a consistent profile of regional brain atrophy on MRI no patient had radiological evidence of significant or strategic vascular damage. Genetic screening of the cohort revealed 11 patients with a pathogenic mutation (six C9orf72, five MAPT). All patients with a genetic mutation presented with bvFTD apart from one patient with a C9orf72 expansion who presented with PNFA.

Patients' care givers completed a semi-structured questionnaire designed to identify symptoms suggesting altered pain or temperature processing (altered experience of pain or temperature) developing since the onset of their illness (table 3.1). This questionnaire recorded care giver descriptions of patients' symptoms and initially sought to capture any unexplained unpleasant physical symptoms more generally, before focussing explicitly on altered behavioural responses to pain or temperature variations. Questionnaire data were analysed to determine the nature of any alteration in pain or temperature responsiveness and its directionality (increased versus decreased), indexing responsiveness from care giver descriptions of patients' verbal and nonverbal output behaviours.

3.3.2 Brain MRI acquisition and analyses:

At the time of questionnaire data collection each patient underwent a volumetric brain MRI scan. In order to assess any relation between individual brain atrophy profile and development of

pain and temperature symptoms, each patient's brain MR scan was reviewed by two experienced cognitive neurologists (PDF, JDW) while blinded to symptomatic and clinical syndromic status. In each case, the presence of any relatively focal brain atrophy (disproportionate to more diffuse background atrophy) and the direction of any cerebral hemispheric asymmetry on visual inspection were recorded for the frontal, temporal and parietal lobes. Pre-processing was performed as per standard methods outlined in chapter 2.

Voxel intensity (grey matter volume) was modelled over the entire patient cohort and within the combined FTLD and AD cohorts, as a function of presence or absence of any symptoms suggestive of altered pain or temperature processing and separately for pain symptoms and for temperature symptoms alone. I conducted separate sub-analyses based on the same model to assess correlates of pain and temperature symptoms within each clinically-defined FTLD syndromic group. In addition, in light of recent evidence suggesting a distinct pathophysiological signature of C9orf72-associated FTLD (Downey *et al.*, 2014; Lee *et al.*, 2014), I performed a sub-analysis of the symptomatic cohort contrasting patients with and without C9orf72 mutations.

Anatomical small volumes based upon prior anatomical hypotheses comprised regions in both cerebral hemispheres critical for pain and temperature processing in the healthy brain; namely, thalamus, and insula (Lenz *et al.*, 1993; Davis *et al.*, 1999; Craig *et al.*, 2000; Brooks *et al.*, 2005; Kim *et al.*, 2007; Isnard *et al.*, 2011; Mazzola *et al.*, 2012). In addition, I assessed symptom correlates in anterior cingulate cortex and within the temporal lobe region anterior to Heschl's gyrus, based on previous work implicating these regions both in appraisal of pain and temperature and other salient stimuli in the healthy brain as well as somatosensory disturbances in disease states (Erickson *et al.*, 2006; Herde *et al.*, 2007; Chan *et al.*, 2009; Cole *et al.*, 2011; Guo *et al.*, 2013). As likely bilateralism of any observed effects could not be predicted a priori, regions of interest were examined for both hemispheres separately. This yielded a total of 8 small volume corrections.

3.4 Results:

3.4.1 Analysis of pain and temperature symptoms:

Characteristics of the patient cohort are summarised in Table 3.2 and a more detailed analysis of symptoms is presented in Table 3.3. The questions presented in the carer questionnaire are given in Table 3.1 and extracts from care giver questionnaire reports for individual patients are presented in Table 3.4.

Symptoms suggesting abnormalities of pain and/or temperature processing were reported in 31/58 patients with FTLD (53% of the FTLD cohort overall) and in 9/20 patients with AD (45% of the AD group). In both FTLD and AD cohorts, altered responses to both temperature and pain variations werereported. While patients with FTLD (13/31 cases, 41%) and AD (3/9, 33%) commonly had altered responses both to pain and temperature, only patients with FTLD (5/31 cases, 16%) had altered pain responses alone. Within the FTLD cohort, symptoms suggesting altered pain or temperature processing were more significantly more frequent in the bvFTD group (15/21 cases, 71%) and SD group (11/17 cases, 65%) than in the PNFA group (5/20 cases, 25%) (bvFTD vs. SD, p=0.65, χ^2= 0.20; bvFTD vs. PNFA, p<0.01, χ^2= 8.84; SD vs. PNFA, p=0.02, χ^2=5.90) accordingly, bvFTD and SD phenotypes were relatively over-represented in the symptomatic FTLD subgroup (Table 3.2).

Has he/she complained of any unusual bodily sensations? If yes, please give details
Has he/she complained of persistent unexplained physical symptoms? If yes, please give details
Does his/her experience of pain seem to have altered compared with before the illness? If yes, please give details
Does his/her tolerance of heat or cold seem to have altered compared with before the illness? If yes, please give details

Table 3.1. Care giver questionnaire to assess pain and temperature symptoms

Characteristic	FTLD: pain / temperature		AD: pain / temperature		Healthy controls	one way ANOVA	two way ANOVA
	Symptoms	No symptoms	Symptoms	No symptoms			
General demographics							
Number: total (F:M)	31(10:21)†	27 (15:12)	9 (2:7)††	11 (6:5)	50 (23:27)		
Number: bvFTD/SD/PNFA	15/11/2005	06/06/2015	NA	NA	NA		
Number: no mutation/C9orf72 MAPT	24/06/2002	23/0/3	NA	NA	NA		
Age (years)	65.4 (52-84)	64.8 (47-80)	63.8 (53-71)	65 (57-74)	67.5 (54-80)		
Education (years)	13.9 (11-20)	15.2 (11-21)	13 (11-17)	15 (12-17)	15.2 (10-18)		
Symptom duration (years)	6.5 (3-21)	4.8 (2-18)	5 (2-8)	5.5 (4-9)	NA		
MRI profile*							
Temporal lobe atrophy (L:R:symm)	22(8:5:9)	16(12:1:3)	6(0:0:6)	9(0:0:9)	NA		
Frontal lobe atrophy (L:R:symm)	10(3:2:5)	11(6:1:4)	0	0	NA		
Parietal lobe atrophy (L:R:symm)	3(1:0:2)	0	2(0:0:2)	1(0:0:1)	NA		
General intellect							
MMSE	**21.1 (4-30)**	**21.9 (1-30)**	**20 (13-25)**	**22.5 (14-29)**	29.6 (28-30)	P<0.0001, r^2= .32, F =9.51	
Verbal IQ	**76 (40-126)**	**78 (55-119)**	**86 (55-115)**	**93 (55-120)**	120 (101-137)	P<0.0001, r^2=0.55, F =38.70	
Performance IQ	**91 (65-136)**	**101 (69-134)**	**81 (59-125)**[a]	**92 (63-119)**	115 (84-141)	P<0.0001, r2= .30, F =13.16	P=0.02, r^2= .08, F =5.53
Episodic memory							
RMT words (/50)	**34 (20-49)**	**37 (18-47)**	**29 (17-42)**	**32 (24-50)**	48 (39-50)	P<0.0001, r^2=0.57, F =42.03	
RMT faces (/50)	**31 (24-50)**[a]	**36 (25-47)**	**32 (18-45)**	**39 (24-46)**	43 (30-50)	P<0.0001, r^2=0.41, F =20.82	P<0.01, r^2= .16, F =6.52
Executive function							
Stroop word (90 sec)	**35 (16-90)**	**39 (18-90)**	**43 (17-79)**	**36.6 (17-58)**	22.7 (15-53)	P=0.0001, r^2=0.19, F =6.42	
Stroop inhibition (180 sec)	**100 (48-180)**	**105 (48-180)**	**143 (42-180)**	**101.4 (30-180)**	57.6 (35-103)	P<0.0001, r^2=0.35, F =13.44	
Digit span reverse (/12)	**3.7 (0-7)**	**4 (0-7)**	**3 (1-6)**	**3.6 (1-7)**	5 (3-7)	P<0.0001, r^2=0.25, F =9.94	

Semantic processing							
BPVS (/150)	**98 (2-149)**	**118 (8-149)**	126 (76-146)	132 (52-147)	147 (137-150)	P<0.0001, r^2=0.26, F =10.56	
Synonyms (/50)	**34 (12-50)**	**38 (20-49)**	**41 (30-49)**	**47.5 (46-49)**	48 (36-50)	P<0.0001, r^2=0.35, F =12.17	
Visuo-spatial							
VOSP (/20)	16 (8-20)	17 (10-20)	**16 (10-18)**	**15 (7-19)**	18 (12-20)	P=0.0002, r^2=0.17, F =6.16	

Table 3.2 Background demographic and neuropsychological information for participants. Mean (range) data are shown unless otherwise indicated and maximum scores on neuropsychology tests are also indicated in parentheses. Initial one-way ANOVA results between all groups (statistical differences relative to controls shown in bold) and then a two-way ANOVA for statistical differences between the FTD and AD groups (a) and within groups for FTD (b). †five patients with altered pain responses only, 13 with altered temperature responses only, 13 with both (see Table 3.3); ††six patients with altered temperature responses only, three with alteration of both pain and temperature responses; *blinded visual rating of brain MRI scans (L:R:symm, number of cases with relatively focal lobar atrophy predominantly left-sided, right-sided or relatively symmetric).

Syndromic diagnosis	**No.**	**MRI profile: focal atrophy***			**Symptom category**	**Response shift**
		TL	**FL**	**PL**		
		L/R/symm	L/R/symm	L/R/symm	P/T/both	inc/dec/both**
bvFTD	15	1/2/5	2/2/3	1/0/0	4/7/4	6/6/3
SD	11	6/2/3	0	0/0/1	1/3/7	8/1/2
PNFA	5	1/1/1	1/0/2	0/0/1	0/3/2	3/0/2
AD	9	0/0/6	0	0/0/2	0/6/3	7/0/2

Table 3.3. Detailed description of the symptomatic patient cohort Key: *blinded visual rating of brain MRI scans (L:R:symm, number of cases with relatively focal lobar atrophy predominantly left-sided, right-sided or relatively symmetric); **variably increased or decreased responsiveness within or between modalities; dec, decreased; FL, frontal lobe atrophy; inc, increased; L, left; P, symptoms of altered pain experience; PL, parietal lobe atrophy; R, right; symm, relatively symmetric; T, symptoms of altered temperature experience; TL, temporal lobe atrophy

Case	Diagnosis	Comment
1	bvFTD	Does not feel pain: has just had three teeth removed, not aware of a problem
2		Seems to have a lot more aches and pains
3		Episodes of fleeting pains in legs over several years
4		Always complains of a big pain in front of head but never complained when skin cancer removed
5		Seems unable to regulate body temperature to the environment; has a fear of the cold
6		Wears jumpers at home and often a dressing gown even in warm weather
7		Complains cold more frequently than before
8		Even in very hot weather will put on a jumper or coat and seems not understand it is hot.
9		Now prefers cold weather
10		Never feels the cold and has to be forced to wear an overcoat
11		Strongly dislikes warm enclosed spaces
12		Does not seem to feel pain as much as before but feels the cold a lot more
13		Fell over and did not complain of pain despite bruises and black eye. Runs baths too hot
14		Last 6 months, has cuts of which seems unaware. Does not feel the cold - wears fewer vests
15		Complains or exclaims more about pains or hurts. Can vary between not feeling the cold to suddenly becoming cold / turning heating up
16	SD	Has a lower pain threshold
17		Feels the cold weather more
18		No longer understands the difference between winter and summer; even in hot weather cuddles a hot water bottle
19		Less tolerant of heat
20		More sensitive to pain. Less able to cope with cold
21		Pains in head, extremely sensitive and exaggerates pain. Always says cold
22		Complains about cuts on his hands and needing more blood. Moans more! Feels the cold more
23		Spurious, short lasting pains all over body, more sensitive to pain. Also complains of itching. Feels the cold more
24		Will make more fuss over small pains. Feels the cold more
25		Pre-illness never went to GP, now goes for minor ailments such as painful thumbs. Less tolerant of cold or heat
26		Difficulty describing the type or severity of pain. Finds cold weather more difficult to cope with; forever commenting the temperature on the car dial
27	PNFA	Needs more heat than before
28		Feels 'cold' almost all the time even on hot day; wraps up while companion in shorts and T shirt
29		Seems unable to tell if too hot or cold; will keep coat on unless told to take it off, walks around in shirt in winter
30		Complains right side of body painful, seems more sensitive to pain. Always cold
31		Now accepts knocks and bumps without comment. More tolerant of both hot and cold
32	AD	Feels the cold more
33		Constantly complains of feeling cold
34		Doesn't seem to be aware when it's hot - will wear too much clothing, seems over-sensitive to cold
35		Feels much colder than before
36		Feels cold a lot more, unable to cope with cold or hot
37		Complains of both hot and cold
38		Complains of pains and feeling the cold a lot more
39		Before pains were not a problem but now are. Always complaining cold
40		More drama around pain. Feels the cold far more, even when well wrapped up, puts heating on all day.

Table 3.4 Care giver comments

Of the genetic FTLD subgroups, patients with C9orf72 mutations were over-represented in the symptomatic subgroup, reporting symptoms suggesting altered pain or temperature processing in all (6/6) cases; whereas these symptoms were recorded less frequently (2/5 cases) for patients with MAPT mutations.

Care giver reports (Table 3.4) revealed a range of phenomenology of altered pain and temperature experience among patients in the symptomatic cohort. Symptoms were of variable intensity and frequency, ranging from mildly to significantly altered. Both increased responsiveness and decreased responsiveness to pain and temperature variations were described, as well as responses that were variably increased or decreased within or between modalities. Within the temperature modality, patients more often developed a dislike of cold (rather than warm) environments. The directional preponderance of altered pain and temperature responsiveness varied between syndromic groups: within the bvFTD group, decreased responsiveness and increased responsiveness to pain and temperature variations were equally frequent (each reported in six cases, 40%); whereas increased responsiveness was more commonly described within the SD group (8/11 cases, 73%), the PNFA group (3/5 cases, 60%) and the AD group (7/9 cases, 78%; see Table 3.2). More complex bidirectional shifts in pain and temperature responses were also described in all syndromic groups.

When patient subgroups within the FTLD and AD cohorts were compared according to the presence or absence of pain and temperature symptoms, the symptomatic FTLD group showed significantly ($p<0.01$, $r2= 0.16$, $t=-2.77$, $F=4.78$) greater impairment of face memory than the non-symptomatic FTLD subgroup; the subgroups within each disease cohort were otherwise similar overall (Table 3.2). Thyroid function (available for 66/78 patients including 34/40 patients with pain and temperature symptoms) was normal in all cases assessed. Three patients in the FTLD cohort with pain and temperature symptoms underwent nerve conduction studies, which were normal in all cases.

Grey matter association	Brain region	Side	Peak (mm)			Z-score	Cluster (voxels)
			x	y	z		
All FTLD	mid insula	R	40	-1	0	4.46	299
	posterior insula	R	38	-18	0	4.02	158
	inferior temporal gyrus	R	62	-9	-30	3.44	115
	superior temporal gyrus	R	62	6	-18	4.14	72
	posterior thalamus	R	15	-31	4	3.66	42
C9orf72 mutations	posterior thalamus	L	-18	-25	1	3.55	120
		R	20	-24	3	3.73	66
Other FTLD	temporal pole	R	52	17	-30	4.59	1026

Table 3.5 Neuroanatomical correlates of altered pain and temperature processing in the FTLD cohort Significant regional grey matter correlates of altered pain and temperature processing (atrophy associated with any symptoms suggesting altered experience of pain and / or temperature) are based on contrasts over the whole frontotemporal lobar degeneration (FTLD) cohort; and in patients with C9orf72 mutations (all symptomatic) versus symptomatic patients without C9orf72 mutations (other FTLD). All associations shown were significant at threshold $p<0.05$ corrected for multiple comparisons within the pre-specified anatomical small volume of interest; all significant clusters >40 voxels are shown and peak (local maximum) coordinates are in Montreal Neurological Institute standard stereotactic space (see also Figure 3.1).

3.4.2 Neuroanatomical correlates of altered pain and temperature processing:

Visual review of individual patient MRI scans (summarised in Tables 3.3) revealed an over-representation of cases with relatively focal and particularly, right-sided temporal lobe atrophy in the subgroup of patients with FTLD and pain and temperature symptoms. Focal temporal lobe atrophy was frequent in all three FTLD syndromes within this symptomatic cohort but concentrated (as anticipated) in the SD subgroup. Disproportionate temporal lobe atrophy was also frequent in patients with AD and pain and temperature symptoms; however, in contrast to the FTLD cases, none of these AD patients exhibited asymmetric temporal lobe involvement nor was there any temporal lobe predilection for symptomatic versus non-symptomatic AD cases.

Regional grey matter correlates of pain and temperature symptoms from the VBM analysis are summarised in Table 3.5 and statistical parametric maps are shown in Figure 3.1. No common grey matter correlates of pain and temperature symptoms were identified for the entire patient cohort (FTLD and AD) at the prescribed corrected significance threshold ($p<0.05_{FWE}$ within the pre-specified anatomical regions of interest). However, within the combined FTLD cohort, the presence of any alteration in pain or temperature responsiveness was significantly associated with atrophy of right anterior superior and inferior temporal cortex, right mid and posterior insula and right posterior thalamus (pulvinar); no significant grey matter associations were identified for pain symptoms or for temperature symptoms in isolation. Pain and temperature symptoms within the AD cohort had no significant grey matter associations at the prescribed corrected significance threshold; however, when statistical parametric maps were examined at a relaxed threshold ($p<0.001$ uncorrected over the whole brain), pain and temperature symptoms in the AD group were found to be associated with grey matter atrophy in the region of the left temporo-parietal junction (local maximum MNI coordinates [-50 – 58 33], Z score 4.13; see Figure 3.2).

In the separate VBM sub-analysis of patients with C9orf72 expansions contrasted with other FTLD patients showing altered pain or temperature responses (Table 3.5 and Figure 3.1), symptoms due to C9orf72 mutations were significantly associated with atrophy of bilateral posterior thalamus (pulvinar) ($p<0.05_{FWE}$ within the pre-specified anatomical region of interest). Conversely, pain and temperature symptoms in the remainder of the FTLD subgroup (directly contrasted with the C9orf72 mutation subgroup) were significantly associated with atrophy of right temporal polar cortex. No grey matter associations were identified within the FTLD syndromic subgroups as clinically defined, at the prescribed significance threshold.

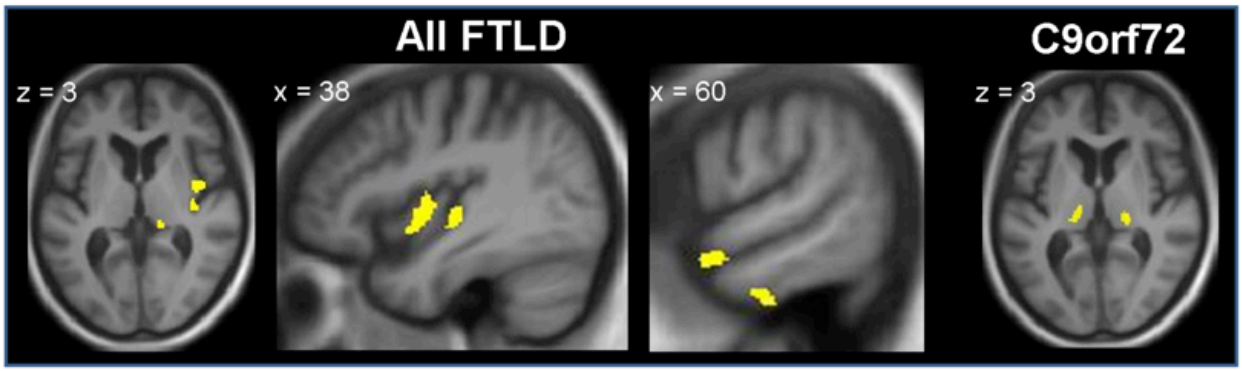

Figure 3.1. SPMs showing regional grey matter atrophy significantly associated with altered pain and/or temperature responsiveness in the FTD cohort. SPMs are based on the contrast between patient subgroups with and without symptoms in the combined (*All FTLD*) cohort and in patients with C9orf72 mutations (*C9orf72*; all symptomatic) versus symptomatic patients without C9orf72 mutations

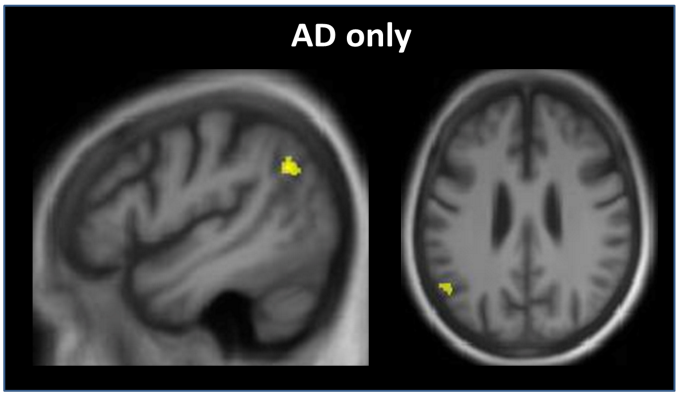

Figure 3.2. SPMs showing regional grey matter atrophy associated with symptoms of altered pain and/or temperature responsiveness within the AD cohort. SPMs are based on the contrast between AD patient subgroups with and without symptoms

3.5 Discussion:

Here I demonstrate that abnormal sensory perceptions are common in FTD and AD and occur with all disease phenotypes but most frequently in the behavioural and semantic variant of FTD. Abnormal perceptions appeared to occur both in the interpretation of one's own internal milieu and of the external sensory environment. They were over-represented in the C9orf72 genetic group. The nature of these symptoms seems to be phenotypically different between the groups with a heightened sensitivity and somatiform behaviour more common in the SD and AD groups and a relative apparent decreased awareness in the bvFTD, which for FTD is in keeping with earlier work (Snowden *et al.*, 2001).

Neuroanatomically, in a combined FTLD cohort, the presence of symptoms was associated with atrophy of three key areas implicated in normal somatosensory processing pathways; the right pulvinar of the thalamus, posterior insula and anterior temporal lobe. Within the symptomatic patients, the thalamic signal appeared to be driven by the genetic C9orf72 cases. These findings substantiate current formulations of the neural organisation of central somatosensory and homeostatic signal processing from previous studies in the healthy brain (Peyron *et al.*, 2000; Craig, 2002; Singer *et al.*, 2004; Henderson *et al.*, 2007; Herde *et al.*, 2007; Craig, 2009; Isnard *et al.*, 2011; Borsook, 2012; Moulton *et al.*, 2012; Preusser *et al.*, 2014)

Given the high frequency of abnormalities of sensory perceptions reported here, these symptoms have received surprisingly little attention in the FTD literature and previous work has not undertaken detailed anatomical correlation. The only neuroanatomical investigation so far has demonstrated that within an SD population unexplained non-specific sensory symptoms appear to be associated with right relative to left ATL atrophy (Bach *et al.*, 2009). That atrophy of the right ATL should relate to impairment in sensory processing in FTD is interesting for several reasons: firstly, the symptomatic and asymptomatic groups were matched for all background neuropsychological measures apart from worse performance on a face recognition memory test in the symptomatic group, a marker of right ATL function. Whilst the left anterior

lobe has long been established to underpin semantic processing of language (Warrington, 1975; Mesulam, 1982) non-linguistic pan-modal semantic deficits, including impairment in extraction of meaning from music, olfaction and non-verbal sound processing occur frequently in FTD and have been associated with right ATL function (Goll *et al.*, 2010; Omar *et al.*, 2010; Hsieh *et al.*, 2011; Downey *et al.*, 2013). In addition, the development of more egocentric behaviour with impairments in interpretation of empathy, morality and theory of mind functions have been localised to the right ATL in FTD (Rankin *et al.*, 2006; Zahn *et al.*, 2009; Irish *et al.*, 2014). The anterior temporal lobe has also been demonstrated as a key hub as part of a more generalised 'appraisal network' (Guo *et al.*, 2013) and therefore, within the pain processing hierarchy, the temporal lobe is likely to play a key role in contextualising unpleasant sensory experience to allow coherent, appropriate behavioural responses; with degradation of this system it is not difficult to extrapolate to how exaggerated, somatising behaviour could arise. Secondly, there is evidence from the epilepsy literature to support a role of the right ATL in the generation of somatic sensations. Although peri-ictal sensory phenomenon are most commonly associated with seizure foci within insular or parietal cortex, sensory phenomenon including perceptions of tingling, numbness, vibrations, burning, prickling and a gnawing type pain have all been reported to occur during temporal lobe seizures, most frequently originating in the right hemisphere (Erickson *et al.*, 2006). Additionally, following right ATL resection for intractable epilepsy, patients have been described who develop de novo abnormal somatic and visceral sensations, including head tingling or a feeling of shrinking, whole body numbness or pains, pins and needles of the legs, or non-specific abnormal cardiac and gastrointestinal sensations (Naga *et al.*, 2004).

In addition to the involvement of the right ATL, I also demonstrated that altered sensory perceptions correlated with grey matter loss in the thalamus and insula; areas well established in pain and somatosensory processing circuitry. Current pain processing models propose that peripheral somatic and visceral sensory afferents relay via thalamic ventromedial nuclei to primary somatosensory cortex and dorsal posterior insula; whilst the latter is implicated in pain

processing (Craig, 2002), primary somatosensory cortex is likely to be more concerned with tactile event processing, perhaps accounting for its absence from the present analysis (Preusser *et al.*, 2014). Lesions of both the thalamus and posterior insula can result in pain, for example, a central pain syndrome characterised by contralateral poorly localised burning pain and paraesthesia is well recognised following strokes of either region (Sprenger *et al.*, 2012). Both insula and thalamus are involved by the pathological process in FTLD (Chow *et al.*, 2008; Zhou *et al.*, 2010; Garibotto *et al.*, 2011); and one may anticipate how abnormal involvement of the ventromedial thalamic nuclei could underlie altered pain and sensory perceptions and therefore, whilst one must cautious about over speculation in studies of this size, it is of interest that it was the pulvinar that we found to be affected here. Abnormal cutaneous temperature perception, numbness, dysasthesias and pain have been reported to occur in a large proportion of patients with new variant CJD (Macleod *et al.*, 2002), a disease that classically devastates the pulvinar relative to other thalamic nuclei. Further, whilst thalamic vascular lesions usually affect multiple nuclei, lesions excluding the VM nuclei can cause pain and in a recent study of post-stroke thalamic pain, anatomically segregating the thalamus using a high resolution 3D atlas, demonstrated that it was pulvinar involvement that led to the highest likelihood of developing a clinical pain syndrome (Kim *et al.*, 2007; Sprenger *et al.*, 2012; Sposato *et al.*, 2014).

A key role of the posterior insula in the cortical processing of pain comes from several lines of evidence: it is the cortical area most consistently activated in functional imaging studies of both visceral and somatic experimentally induced pain paradigms (See (Peyron *et al.*, 2000) for a meta-analysis); human lesions result in pain (as well as thermanaesthesia and anaesthesia), (Greenspan & Winfield, 1992; Schmahmann & Leifer, 1992; Isnard *et al.*, 2011) and a recent large analysis of micro-stimulations made over the whole cortex as part of pre-surgical epilepsy evaluations revealed that the posterior insula was the *only* cortical region where stimulation produced pain (Mazzola *et al.*, 2009; Mazzola *et al.*, 2012). However, the VM thalamus and posterior insula have roles in sensory processing beyond that purely of pain

analysis; like other areas involved in sensory processing, both thalamic and posterior insula stimulation in awake humans can also evoke can evoke a variety of non-painful sensory perceptions including feelings of cooling, tingling, vibration, numbness, pins and needles and electrical sensations (Lenz *et al.*, 1993; Lenz *et al.*, 1994; Davis *et al.*, 1999; Mazzola *et al.*, 2009; Mazzola *et al.*, 2012), the posterior insula shows functional activation with sensations of warming and cooling, itch, sensual touch and those arising from the viscera (for example, breathlessness), (King *et al.*, 1999; Banzett *et al.*, 2000; Craig *et al.*, 2000; Olausson *et al.*, 2002). Activity encodes stimulus intensity (Craig *et al.*, 2000) and is somatotopically organised in both thalamus and insula (Brooks *et al.*, 2005; Hua *et al.*, 2005; Henderson *et al.*, 2007; Bjornsdotter *et al.*, 2009). The posterior insula has therefore been proposed as a sensory hub involved in integration of these homeostatic signals to map an interoceptive 'image' of body state (Craig, 2002). It projects to the anterior insula via strong projections through the middle insula, which receives further sensory input such as gustatory and vestibular information (Small, 2010) and whereas functional activity on imaging studies correlates with stimulus strength in the posterior insula, activity better correlates with *perceived* stimulus intensity in the anterior insula and is modulated by psychological factors (Craig *et al.*, 2000) (Brooks *et al.*, 2002). The anterior insula is a hub of sensory, autonomic and perceptual integration, reflecting conscious awareness of one's own internal state, and even abstract concepts such as time perception (Critchley *et al.*, 2000a; Critchley, 2005; Seeley *et al.*, 2007a; Seeley *et al.*, 2007b; Corbetta *et al.*, 2008; Sridharan *et al.*, 2008; Seeley *et al.*, 2009; Menon & Uddin, 2010; Critchley *et al.*, 2011; Beissner *et al.*, 2013; Mueller-Pfeiffer *et al.*, 2014). It has therefore been proposed that whilst the posterior insula processes information on what where and how strong a stimulus is, and the middle insula is involved in further cross-modal sensory integration, the anterior insula acts to allow awareness of one's bodily state and the subjective feelings of self (Craig *et al.*, 2000; Craig, 2002; 2009). I therefore suggest here that abnormal sensory symptoms in FTD may arise secondarily to a breakdown of the normal thalamo-posterior insula network, resulting in disrupted topographic and somatotopic information reaching the anterior insula.

Whilst the number in this study in whom genetic testing had been performed is small, it is of interest that all of the C9orf72 cases showed sensory symptom abnormalities, in contrast to the MAPT cases, where incidence was the same as the rest of the cohort. That mutation in the C9orf72 gene underlies FTD phenotypes is a recent discovery and the range of phenotypes and anatomical signatures resulting from C9orf72 mutations is wide and requires further investigation (Mahoney *et al.*, 2012b; Snowden *et al.*, 2012; Whitwell *et al.*, 2012). However, it is emerging that psychiatric symptoms and disruption of body schema perceptions appear to be early and pervasive features (Downey *et al.*, 2012a; Snowden *et al.*, 2012; Takada & Sha, 2012; Snowden *et al.*, 2013). Further, abnormal perception of pain has recently been described in a series of patients with C9orf72 mutations in which 7 of 12 exhibited diffuse pain symptoms of the head, abdomen, chest and legs. Headaches and abdominal pains appeared to be the most frequent (5 of 7) and in one report began in the patient's 30s, pre-dated the evolution of behavioural symptoms by more than 30 years. Pathology was available on 4 of these cases and was heaviest in the frontal and temporal cortices and the thalamus (Landqvist Waldo *et al.*, 2013).

The VBM analysis of the AD cohort here revealed (at a more lenient statistical threshold) a distinct cortical correlate of pain and temperature symptoms in the region of the temporo-parietal junction. While interpretation of such uncorrected data must be cautious, this cortical region has been implicated in processing pain and in particular, in reorienting brain activity between resting 'default mode' and active attentional modes in response to salient stimuli (Kucyi *et al.*, 2012a; Bray *et al.*, 2014). The temporo-parietal junction is a core target of pathology in AD (Warren *et al.*, 2012) and may be involved in a range of behavioural features in this disease that presently remain less well characterised than the behavioural syndrome of FTLD. These AD-associated behavioural changes include anxiety and hyper-emotionality, features that also typically develop in chronic pain syndromes (Sturm *et al.*, 2013; Kucyi & Davis, 2014; Pujol *et al.*, 2014). Aberrant activity of temporo-parietal cortex in AD might disrupt processing of interoceptive signals, both by amplifying self-reflective awareness of body states

via the default mode network and by increasing the salience of signals via the salience network (Grecucci *et al.*, 2013; Letzen *et al.*, 2013; Zhou & Seeley, 2014). The relative prominence of temperature (relative to pain) symptoms in the AD cohort here supports this interpretation, since under most circumstances thermal comfort or distress reflects the degree of perceived mismatch between one's own body temperature and the environment (Craig, 2002).

Based upon these findings I suggest a model for outlining potential neurodegenerative disease effects on these processes that is consistent with data from the healthy brain and the effects of focal brain lesions (Craig, 2002; 2009; Borsook, 2012) (Figure 3.3). According to this model, C9orf72 mutations target thalamic gated sensory coding; while bvFTD more generally disrupts the relay of body state information from posterior insula and its further visceral and somatosensory integration in mid insula to more anterior regions, resulting in perceptual constructs that are based upon distorted 'noisy' afferent sensory signals allowing for either abnormally reduced or abnormally increased subjective awareness of homeostatic signals, (a mechanism potentially analogous to pain asymbolia following focal insular lesions (Berthier *et al.*, 1988; Masson *et al.*, 1991)). Degeneration of anterior temporal lobe mechanisms in SD impairs contextual processing of homeostatic information via the insula, resulting in 'over-valuation' (decreased tolerance) of such stimuli; while temporo-parietal cortical damage in AD leads to aberrant salience coding of homeostatic signals via interruption of normal SN and DMN interactions (Zhou & Seeley, 2014).

From a clinical perspective, the present work provides a framework for understanding an important category of symptoms that have received relatively little attention in neurodegenerative disease. My findings underline the prevalence of pain and temperature symptoms across the FTLD spectrum. The syndrome associations may underlie the somatisation, hypochondriasis and abnormal illness behaviour that these patients frequently exhibit, particularly in the setting of focal right temporal lobe atrophy (Snowden *et al.*, 2001; Chan *et al.*, 2009) where impairment of contextual meaning might drive such behaviours. Perhaps more surprisingly, my findings suggest that similar symptoms (particularly affecting

thermoregulatory signals) are not uncommon in patients with AD and may have been under-recognised.

VBM findings must be interpreted with a degree of caution; the use of a multiple regions of interest statistical approach, raises the possibility of false positive errors. Whilst the use of a whole brain statistical approach restricts false positive error, this of course inevitably incurs a corresponding potential cost in false negatives. When dealing with cognitive systems where there is ample evidence to suggest, a priori, that many brain regions are not functionally relevant to the system under investigation, use of a whole brain analysis statistical approach is inappropriately stringent and the use of a region of interest approach is generally considered in the literature to be the most appropriate (ref). Post-hoc analysis of a region of interest based approach using a simple Bonferroni correction for number of tests would also be inappropriately harsh, as these are not truly independent analyses (it is likely a priori that the areas affected represent functionally interconnected regions). I therefore also analysed the VBM data using a single combined, bi-hemispheric thalamic and insula region (rather than two separate hemispheric regions, as previously) for the small volume correction for the combined cohort analysis. The results (not shown here) as anticipated were attenuated but substantially unaltered and continue to show significant correlations in the pre-specified anatomical regions of interest. Therefore the results are likely robust, especially in light of their congruity with areas previously shown to be involved in these processes. The sample sizes here are small in relation to those used in other studies and replication of this work in future studies with larger patient groups (and more detailed correlations) would allow for stronger statistical strength and shed light on this issue.

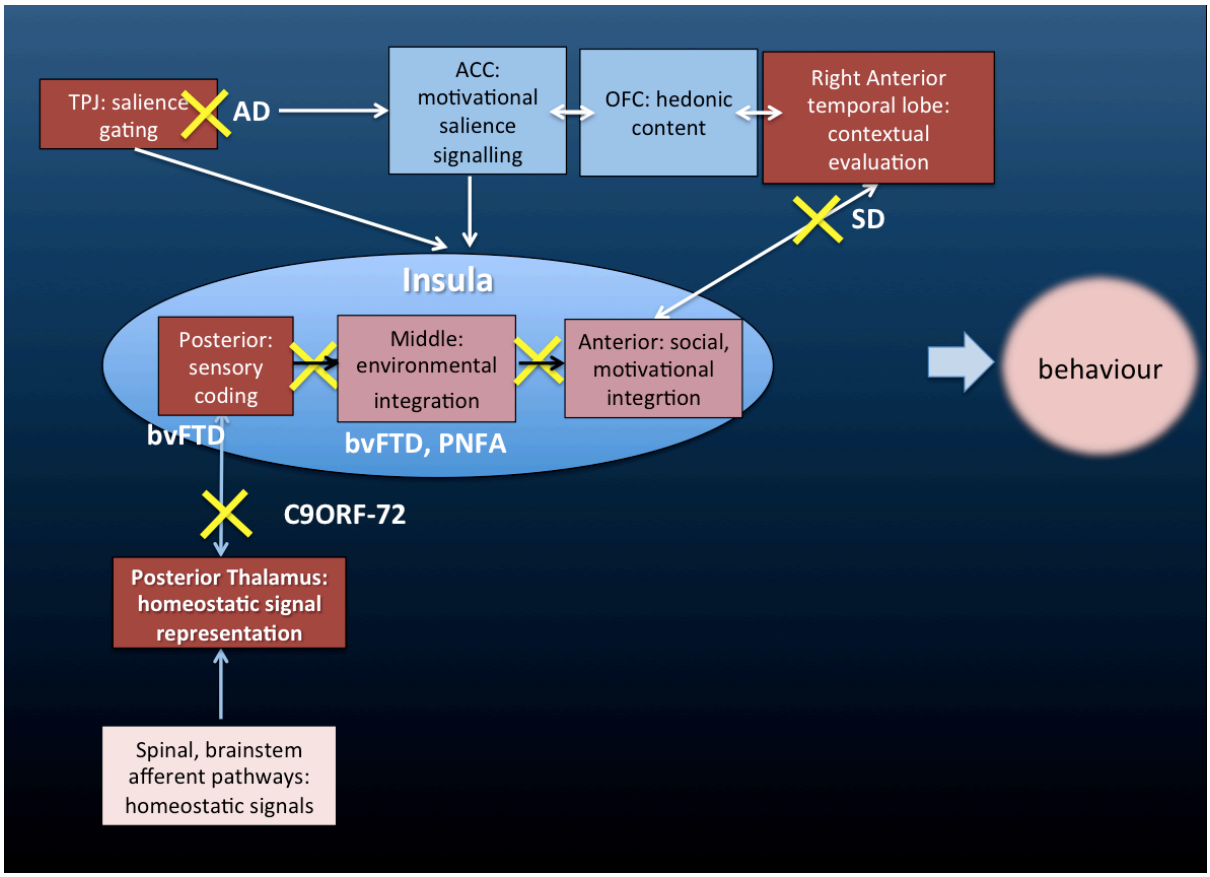

Figure 3.3. A schematic synthesis of the effects of dementia syndromes on pain and temperature processing, based on present data and current formulations of central homeostasis (Craig, 2002; 2009; Borsook, 2012; Zhou & Seeley, 2014). Ellipses indicate core components of the homeostatic processing network, oblongs indicate linked regions that modulate processing of homeostatic signals and arrows signify predominant direction of information flow; anatomical regions are labelled above their putative roles in the processing hierarchy and dementia syndromes are labelled (italics) alongside grey crosses indicating the major locus of dysfunction in that syndrome. According to the proposed synthesis, C9orf72 mutations target early encoding of pain and temperature signals in thalamo-cortical circuitry; bvFTD disrupts the relay of body state information from posterior insula and both bvFTD and PNFA degrade its contextual integration in mid insula and more anterior regions; SD degrades anterior temporal lobe mechanisms that evaluate stimulus context; and temporo-parietal cortical damage in AD may lead to abnormally enhanced gating and aberrant salience coding of homeostatic signals.

3.6 Chapter Conclusions:

Here I have demonstrated that abnormalities in sensory processing are a common in FTD, and mainly reflect a disruption of pain and temperature processing. The nature of these symptoms seems to be phenotypically different between the groups with heightened sensitivity behaviour more common in the SD and AD groups, and a relative decreased awareness in bvFTD. Neuroanatomical correlations revealed that these symptoms are underpinned by damage to key areas in brain somatosensory processing pathways: the pulvinar of the thalamus, the posterior insula cortex, and the right anterior temporal lobe in FTD and the temporo-parietal junction in AD. The C9orf72 mutation cases likely drove the pulvinar signal observed and it is probable that the different disease groups maximally target different aspects of a normal processing hierarchy, partially accounting for different phenotypes seen in the different groups. These findings add to the evidence that symptoms common to FTD (and additionally AD) reflect distortions of core sensory coding, salience assignment and contextual placement.

Chapter 4: Altered physiological reactivity to primitive salience cues in FTD

4.1 Chapter summary:

Abnormal responsiveness to salient sensory signals may underlie behavioural changes observed in FTD, but has been little studied. In healthy older adults approaching sounds have been demonstrated to evoke larger autonomic reactions than their withdrawing counterparts. Here I manipulated tones using intensity cues to create perception of salient approaching ('looming') or less salient withdrawing sounds and pupillary and behavioural rating responses to these stimuli were compared in patients with FTD and AD and a cohort of healthy age-matched individuals in order to investigate whether these normal differential responses were deranged in FTD and AD. Approaching sounds were rated as more salient than withdrawing sounds by healthy older individuals but this behavioural response to salience was not demonstrated in dementia syndromes. Overall pupil reactivity to both approaching and withdrawing sounds was decreased in patients with C9orf72 mutations relative to healthy older controls and other patients with bvFTD. Pupillary responses to approaching sounds were greater than responses to withdrawing sounds in healthy older individuals. However, in contrast to the a priori hypothesis that this differential effect would be lost in SD and preserved in PNFA, the differential pupil response was exaggerated in SD relative to healthy controls and significantly depressed in patients with PNFA and AD relative both to the healthy control and SD groups. This suggests that auditory salience coding from perceived motion direction is more posteriorly encoded and may be differentially affected by dementias. This may provide a starting point for development of novel physiological biomarker of these diseases.

4.2 Introduction:

Stimuli that appear to approach or 'loom' are more salient than those that appear to recede and the potential evolutionary role of this is clear; the ability to preferentially shift attention towards stimuli that are approaching carries a survival advantage, both for engaging with desirable stimuli and avoiding those that pose a threat. Although this effect is best demonstrated with full motion cues, it can also be evoked with intensity shifts alone, suggesting that much salience information is carried in low-level perceptual cues (Neuhoff, 1998; Seifritz *et al.*, 2002; Bach *et al.*, 2008).

This preferential responsiveness to approaching versus withdrawing stimuli has been demonstrated in both human and non-human primates. Monkeys orientate for longer towards visually enlarging relative to shrinking shapes and to the source of increasing rather than decreasing intensity sounds (Schiff *et al.*, 1962; Ghazanfar *et al.*, 2002). In humans, thresholds for detection of intensity differences between simple static short tones are lower when the comparator tone is of greater rather than lesser intensity than the baseline tone (Ellermeier, 1996). For dynamic sounds, relative to those decreasing in intensity, increasing intensity sounds are estimated as being closer, louder, appear to be moving faster and are overall perceived as more unpleasant, alerting and threatening (Ellermeier, 1996; Grose & Hall, 1997; Neuhoff, 1998; Stecker & Hafter, 2000; Bach *et al.*, 2008; Bach *et al.*, 2009; Cappe *et al.*, 2009) and it has been suggested that an overestimation of their speed and proximity of approaching sounds may together have provided an evolutionary advantage allowing a margin of error in which to shift attentional resources to relevant stimuli before contact (Popper & Fay, 1997).

Using 2 second 1Khz carrier frequency pure tones, Bach et al. (2008), presented young healthy adults with rising, falling and constant intensity tones whilst measuring changes in SCR and HR as metrics of autonomic system reactivity. As well as being rated as more alerting, approaching sounds produced consistently greater autonomic responses than withdrawing sounds, consistent with those produced in an orienting response (Raskin *et al.*, 1969). In a concomitant fMRI paradigm the authors also demonstrated functional activation of the

amygdala, left temporal plane including the superior temporal sulcus (STS) and proposed that the amygdala is acting here as a 'warning cue'. Two further studies have confirmed anatomical areas implicated in these preferential responses include cortical regions within the right hemisphere associated with phasic attention (for example the STS/TPJ) and left hemisphere areas of the temporal plane, (implicated in spatial analysis and motion signalling, (Griffiths & Warren, 2002; Goll *et al.*, 2012)). The STS is implicated in cross modal integration (Beauchamp *et al.*, 2004; Noesselt *et al.*, 2010; Werner & Noppeney, 2010) and has shown activation in studies using just auditory as well as mixed visual and auditory looming stimuli (Seifritz *et al.*, 2002; Bach *et al.*, 2008; Tyll *et al.*, 2013). It receives input from visual and auditory regions (Seltzer & Pandya, 1994; Padberg *et al.*, 2003) and seems to play a role in the processing of audio-visual motion stimuli in general (Baumann & Greenlee, 2007; Werner & Noppeney, 2011). The TPJ (in a region that overlaps with the posterior STS in the study by Bach et al., 2008) likely has a role in attention and salience signalling; not only does it respond to novel and salient stimuli (Downar *et al.*, 2000; 2001; 2002; Downar *et al.*, 2003), with resting state fMRI it has recently been demonstrated to connect strongly to a network that includes areas key to the salience network as outlined in chapter 1 (Kucyi *et al.*, 2012a). However, although two further functional imaging studies investigating the neural substrate of the processing of looming sounds have shown similar activity in the dorsal regions (STS, TPJ), they did not show similar amygdala activity as demonstrated by Bach et al (Seifritz *et al.*, 2002; Tyll *et al.*, 2013). The amygdala has been long established as a key hub as part of the limbic system of emotional processing, most commonly in experiments of fear conditioning. Activity has been shown in response to subconsciously perceived threat images and it is postulated that its role here is relevant in signalling a potentially hazardous stimulus as a 'warning cue' for rapid alerting to sources of threat, without the need for conscious appraisal (Liddell *et al.*, 2005; Bach *et al.*, 2008). The amygdala is a functionally complex and heterogeneous structure and remains incompletely defined. In addition to assignment of attention, emotion and reinforcement learning as discussed above, amygdala responses have also been shown to respond to stimulus

properties that include novelty (Zald, 2003), arousal (Whalen *et al.*, 1998; Lewis & Barton, 2006), relevance (Sander *et al.*, 2003; Sander, 2012), and ambiguity (Rosen & Donley, 2006).

Neurodegenerative diseases differentially target key components of the networks involved in processing the psychological and physiological aspects of looming, and from first principles, one could speculate that this normal psychological and physiological bias towards looming will be altered. If the amygdala is a key component driving response, then the diseases AD and SD, that show particular involvement of the amygdala, should display particularly derangement of these responses (either psychological, physiological or both).

Hypotheses and predictions: Here I hypothesised that normal preferential responses towards approaching sounds would be deranged in FTD reflecting deranged salience processing. More specifically, I predicted that, as this has previously been linked with amygdala function, SD, showing greater amygdala damage relative to the other syndromes, would demonstrate loss of this effect and that PNFA, with the least amygdala damage would likely show normal responses; these differences in response between FTD groups could then be used to parse these syndromes on a physiological level.

4.3 Methods:

4.3.1 Participant characteristics:

10 SD, 16 bvFTD, 12 PNFA, 10 AD patients and 26 healthy age matched controls participated in the pupillometry experiment (see for baseline demographics). Twelve patients with bvFTD had proven genetic mutations (six C9orf72, six MAPT). The study groups did not differ in mean age and patient groups did not differ for mean symptom duration; there were more males in the bvFTD group.

4.3.2 Pupillometry experiment:

Files were synthesised to produce digital sounds perceived as approaching, Iup (tones with increasing intensity) versus withdrawing, Idown (tones with decreasing intensity). Carrier

sound stimuli were synthesised as pure tone wavefiles under Matlab 7.0® (http://www.mathworks.co.uk/) at base frequency 700 or 1000Hz as narrow-band sounds in this frequency range have been shown previously to evoke robust behavioural and physiological responses to auditory looming in the healthy brain (Neuhoff, 1998; Seifritz *et al.*, 2002; Bach *et al.*, 2008; Bach *et al.*, 2009). In keeping with earlier work (Bach *et al.*, 2008), all tones were 2 seconds duration with the same base mean intensity (root mean square) level. Intensity changes were applied as linear ramps between 0 and 75dB with 5ms onset and offset ramps to eliminate click artefacts. These large intensity changes were easily perceived by all participants and sounds with increasing or decreasing intensity were reported by the healthy older controls to generate a percept of 'approaching' or 'withdrawing', respectively. The experiment was designed such that 10 synthetic sounds were presented in a pseudo-randomised order interspersed with a playlist of 30 familiar nonverbal sounds (representing common human vocal, animal, mechanical and environmental noises), such that every fourth sound was artificial. The interspersion of sounds was intended to prevent habituation or anticipation of a particular stimulus type. To help maintain variety, 4 additional artificial sounds (including two with 400Hz carrier frequencies) were included. These sounds were excluded from the final analysis as sounds of this carrier frequency have not been shown to evoke consistent pupillary reactions in previous work.

During each trial there was an initial brief silent interval (two seconds), followed by the sound stimulus (two seconds) and a final silent equilibration interval (nine seconds).

4.3.3. Analysis of behavioural and physiological data:

All data points were included in the analysis using the random effects statistical method to account for non-independence as described in section 2.2.3. As males were over-represented in the bvFTD group, gender was incorporated as a nuisance covariate. Alerting ratings were not included in this experiment as stimuli were matched in intensity and approaching sounds are known to be perceived as louder and more alerting. When comparing whether the magnitude of

difference in responses to approaching versus withdrawing sounds differed between groups, a single 'difference' measure for each individual was calculated by subtracting the mean response to withdrawing sounds from that to approaching sounds. Group differences were then compared with an initial one-way ANOVA and then linear regression comparisons between each group.

4.4 Results:

4.4.1 General characteristics of participant groups:

Demographic, clinical and general neuropsychological data are summarised in Table 4.1; Participant groups did not differ in mean age or in mean symptom duration. Baseline peripheral hearing thresholds did not vary between patient groups.

4.4.2 Behavioural ratings:

Behavioural alerting rating and pupil response data are summarised in Figure 4.1. The PNFA group rated approaching sounds as significantly less alerting than controls (p=0.02, z=-2.43, χ^2= 10.02); there were no other group differences in mean overall alerting ratings for individual sound conditions. The healthy control group rated approaching sounds as significantly more alerting than withdrawing sounds (p=0.04, z=-3.12, χ^2= 4.12). However, no patient syndromic group or genetic subgroup showed a significant mean difference in alerting ratings between the two sound conditions. Creating a difference measure for alerting responses to approaching minus withdrawing sounds for each individual, a one-way ANOVA demonstrated that there were no group differences in magnitude of the differential responses.

	Controls	SD	FTD	PNFA	AD	One way ANOVA
General						
No.	26	10*	14	12**	10*	
Gender distribution (f:m)	12:14	6:4	3:11	3:9	5:5	
Age (yrs): mean (range)	67 (57-74)	65 (56-78)	66 (52-84)	68 (57-79)	66 (60-78)	
Education (yrs)	16.6 (2.0)	15.0 (3.2)	14.6 (3.4)	15 (3.1)	15.3 (2.4)	
Symptom duration (yrs)	NA	4.5 (2.1)	8.3 (6.2)	4.3 (2.1)	5.3 (2.1)	
IQ						
Verbal	123 (8.2)	**81 (17)**	**89 (20)**	**77 (15)**	**101 (14)**	P<0.0001, r^2= 0.67, F= 30.86
Performance	119 (14)	**111 (16)**	**97 (17)**	**98 (17)**	**90 (16)**	P<0.0001, r^2= 0.41, F= 10.27
Episodic memory						
RMT words (/50)	47 (3)	**30 (8)**	**35 (6)**	40 (8)	**30 (5)**	P<0.0001, r^2= 0.61, F= 22.52
RMT faces (/50)	44 (4)	**36 (8)**	**34 (6)**	**38 (5)**	**32 (5)**	P<0.0001, r^2= 0.46, F= 10.56
Semantic processing						
BPVS (/150)	148 (2)	**103 (45)**	**132 (15)**	**132 (24)**	**140 (8)**	P<0.0001, r^2= 0.53, F= 16.32
SMT score (/45)	40 (5.2)	**35 (10.9)**	**35 (8.1)**	38 (6.2)	38 (7.1)	P<0.0001, r^2= 0.44, F= 10.16
Executive function						
D-KEFS Stroop word	21 (4)	26 (9)	**27 (9)**	**50 (14)**	**31 (9)**	
D-KEFS Stroop inhibition	57 (16)	77 (34)	**94 (42)**	**118 (51)**	**116 (47)**	P<0.0001, r^2= 0.63, F= 15.17
Digit span reverse (max)	5 (1)	5 (2)	5 (1)	**3 (1)**	5 (2)	P=0.0002, r^2= 0.38, F= 7.05
Visuospatial						
VOSP (/20)	18 (2)	16 (3)	17 (2)	16 (2)	**16 (2)**	P=0.0004, r^2= 0.34, F= 6.32

Table 4.1. Demographic, clinical and general neuropsychological data for all participant groups: Maximum scores on neuropsychological tests are shown in parentheses; mean (standard deviation) data are shown unless otherwise indicated. Initial one-way ANOVA results between groups where statistically significant group differences ($p<0.05$) are shown in the right column and significant group differences in patients relative to healthy controls are in bold. *general neuropsychological data in 9 patients **general neuropsychological data in 10 patients; †experimental nonverbal auditory semantic matching test (see text)

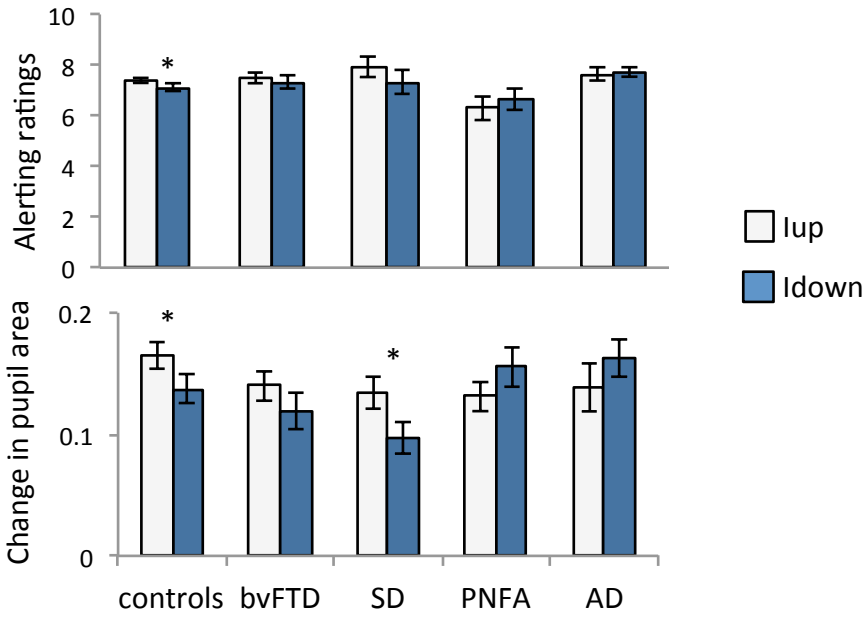

Figure 4.1. Mean alerting ratings (upper panel) and maximal pupil responses (lower panel) for the experimental groups for approaching (intensity increasing, **Iup,** light grey) and withdrawing (intensity decreasing, **Idown,** blue) sound conditions. Alerting ratings are on a Likert scale (1, not all alerting; 10, highly alerting) and pupil responses are shown as log percentage maximal area change from baseline ($Pupil_{max}$). Mean values are shown (error bars signify 1 standard error). * indicate significantly ($p<0.05$) greater responses to approaching than withdrawing sounds

4.4.3 Pupillometric data:

Comparing pupillary responses between groups, overall pupil reactivity (i.e. mean pupil response to sounds irrespective of sound condition) did not differ between syndromic groups. However, there was an overall difference when comparing all genetically divided groups (one-way ANOVA, $p<0.001$, $F=4.44$, $r^2=0.1$,); the bvFTD subgroup with C9orf72 mutations had significantly depressed overall pupil responses relative both to healthy controls and to patients with sporadic bvFTD (C9orf72 vs. controls; $p=0.01$, $z=-2.58$, $\chi^2=9.87$; C9orf72 vs. sporadic bvFTD; $p< 0.01$, $z=-2.6$, $\chi^2=10.47$) and borderline depressed responses relative to the subgroup with MAPT mutations ($p=0.05$, $z=2.8$ $\chi^2=7.82$) but no significant difference relative to the SD, PNFA and AD groups.

In the healthy control group, approaching sounds evoked significantly greater pupil dilatation ($p<0.01$, $z=-2.73$, $\chi^2= 7.48$) than withdrawing sounds. The SD group (but no other syndromic group or genetic subgroup) retained the normal profile of significantly greater pupillary responses to approaching than withdrawing sounds ($p=0.02$, $z=-2.27$, $\chi^2= 5.14$).

An overall difference in response measure was calculated for each individual (their average response to approaching sounds minus their average response to withdrawing sounds); there was an overall difference in the magnitude of these differential responses between groups (one-way ANOVA, $p=0.02$, $F=3.07$, $r^2=0.15$) with responses smaller in both the PNFA and AD groups relative to, firstly, healthy older controls relative to the healthy control (PNFA vs. controls, $p<0.01$, $F=1.98$, $r^2=0.15$, $t=-2.83$; AD vs. controls, $p=0.03$, $F=1.98$, $r^2=0.15$, $t=-2.18$) and secondly, the SD group (PNFA vs. SD, $p=0.01$, $F=4.04$, $r^2=0.22$, $t=-2.69$; SD vs. AD $p=0.04$, $F=4.04$, $r^2=0.22$, $t=-2.16$) but not relative to the bvFTD group.

There were no significant correlations between pupil response and disease duration, overall disease severity (as indexed by MMSE), general semantic (BPVS) or nonverbal auditory semantic (sound classification test) performance over the patient cohort.

4.5 Discussion:

Here I show that approaching sounds are rated as more alerting, and evoke greater physiological responses than their withdrawing counterparts as measured by pupillometry in healthy older adults, consistent with findings in young healthy controls as indexed with changes in SCR in previous work (Bach *et al.*, 2008). They therefore determine that pupillometry is a suitable experimental tool for measuring autonomic function in dementia patients.

Within the dementia syndromes, physiological profiles were differentially disrupted; patients with SD (like healthy individuals) showed greater pupil dilatation in response to looming sounds than withdrawing sounds, whereas this salience signal was lost (relative both to healthy controls and patients with SD) in PNFA and AD. This is particularly intriguing in SD, considering that despite impairment in the ability to classify sounds explicitly (as demonstrated by their impaired performance on the semantic matching task), the physiological salience response to sound was preserved; physiological markers (in SD) may continue to signal the salience of sensory stimuli even where explicit evaluation is disrupted. The findings further suggest that physiological signatures may stratify dementia syndromes.

That the magnitude of the difference in responses to the two sound conditions was significantly reduced in the PNFA and AD groups relative to the normal responses demonstrated in the healthy older controls and the SD group is interesting for several reasons. Firstly, anterior and mesial temporal structures (such as amygdala) that evaluate the behavioural and emotional salience of sensory stimuli (Bach *et al.*, 2008) are typically heavily involved by the pathological process in SD (and were involved on MRI in all cases here) but less consistently damaged in other dementia syndromes; in contrast, posterior temporo-parietal circuitry implicated in salience processing in the context of low level auditory perceptual and spatial analysis (focused upon the TPJ) (Downar *et al.*, 2000; Seifritz *et al.*, 2002; Bach *et al.*, 2008; Kucyi *et al.*, 2012a; Kucyi *et al.*, 2012b) are typically involved in AD and PNFA but relatively spared in SD (Grossman, 2012; Warren *et al.*, 2012; Warren *et al.*, 2013b). Though any

anatomical conclusion here must be tentative, the findings suggest that salience derived from looming relies more upon low-level perceptual characteristics rather than amygdala limbic mediated emotional responses but leave open the critical locus for generating evaluative behavioural responses (which were impaired across dementia syndromes here). Secondly, this simple auditory metric can potentially physiologically parse out patients by clinical syndrome (depressed in PNFA and AD relative to SD and healthy older controls) and within a given clinical phenotype by molecular substrate (significantly depressed in patients with C9orf72 mutations relative to non-genetic bvFTD patients and healthy older adults). This molecular stratification may help to resolve certain apparent inconsistencies with respect to autonomic reactivity profiles attributed to combined bvFTD syndromic cohorts in previous work: baseline autonomic (skin conductance) reactivity has been reported to be decreased in bvFTD (Robles *et al.*, 1999; Joshi *et al.*, 2014; Struhal *et al.*, 2014), while autonomic responses to salient sounds have been reported to be depressed (Hoefer *et al.*, 2008) or retained (Sturm *et al.*, 2006). Physiological signatures may be particularly pertinent where diseases overlap clinically and anatomically (for example, sporadic and C9orf72 driven bvFTD) or where differentiation of molecular pathologies is currently difficult due to convergent phenotypic effects, in particular bvFTD (Warren *et al.*, 2013a; Warren *et al.*, 2013b).

Although differences were only demonstrated on a group level, and one must remain reticent about over extrapolating results based on small groups to individual responses, this does suggest potential clinically relevant avenues in the future. Whilst all the subjects were fairly established in their clinical disease courses, it may be that such differences in physiological phenotypes occur early in disease processes and may offer avenues for clarifying ambiguous early clinical phenotypes. For example, the significantly reduced responses shown in the AD group relative to the healthy control group may have a role in better predicting of those with early subjective memory complaints who may progress to clinical AD. Equally, from a trial perspective, detection of early physiological changes may allow appropriate timing of disease

modifying treatments and tracking responses from a stage where neuroimaging or standard psychometric measures remain insensitive.

This study has several limitations that suggest directions for future work. Case numbers were small, and patients were studied within a relatively limited window of clinically established disease and without direct neuroanatomical or pathological substantiation. Moreover, I adopted a single model physiological paradigm: pupillometry of salient sounds. Further work should evaluate physiological response profiles in larger cohorts, longitudinally over the course of disease (including presymptomatic, genetically at-risk cases), with other physiological metrics and in different sensory modalities, and with neuroanatomical and (ultimately) histopathological correlation.

4.6 Chapter conclusions:

The current data demonstrate that normal physiological bias towards the greater salience of perceived approaching motion is preserved in SD but lost in PNFA and AD dementia types. That this response was lost in those dementia types with more posterior cortical involvement supports the notion that salience extraction from simple motion cues resides more with TPJ structures. Clinically, dementias could be separated both on a syndromic and molecular level, which provided potential avenues for further exploration in the future.

Chapter 5: A disruption of more complex emotional salience processing

5.1 Chapter summary:

Emotional behavioural disturbances are frequent in fronto-temporal dementias but their pathophysiology is poorly understood. Key anatomical regions in emotional processing, such as the amygdala have been implicated as well as disruption to larger neuronal networks, in particular the reciprocal actions of salience and default mode network functions (Zhou & Seeley, 2014). Here I measure pupillary reactions in response to emotional sounds to better define the underpinning of these behaviours and to determine whether separate physiological signatures can be obtained for different dementia syndromes. Work in healthy young controls suggests that highly valent stimuli should evoke greater autonomic reactions, as measured by SCR and HR (Bradley & Lang, 2000; Gomez & Danuser, 2004). Pupil responses and affective valence ratings for nonverbal sounds of varying emotional salience were assessed in patients with bvFTD, SD, PNFA and AD versus healthy age-matched controls.

Results showed that relative to healthy individuals, overall autonomic reactivity to sound was normal in Alzheimer's disease but reduced in other syndromes, including PNFA. As predicted, the coupling between affective behavioural response (valence rating) and pupillary response was preserved in patients with PNFA but deranged in bvFTD, SD syndromes. Slightly surprisingly, responses were also deranged in AD. In SD the relationship between valence of sound and subjective rating returned when measured against the group's ratings rather than the normal pleasantness of a sound as determined by the healthy controls, supporting the hypothesis that derangement in this group may at least partially reflect impaired semantic processing. Overall, different autonomic profiles to affective sounds in dementias may help

better define pathophysiological mechanisms and provide potential for the evolution of new physiological biomarkers.

5.2 Introduction:

Behaviours suggestive of abnormal emotional processing are common in both FTLD and AD, although probably under-recognised. They are particularly pronounced and an early feature in behavioural variant frontotemporal dementia (bvFTD) but also occur in SD (Snowden *et al.*, 2001; Rosen *et al.*, 2006; Snowden *et al.*, 2008; Bediou *et al.*, 2009; Rascovsky *et al.*, 2011; Kumfor & Piguet, 2012; Perry *et al.*, 2014; Zhou & Seeley, 2014). Deficits in emotional recognition have also been demonstrated in PNFA, albeit to a lesser extent (Rohrer & Warren, 2010a) and Alzheimer's disease (AD) (Verdon *et al.*, 2007; Bediou *et al.*, 2009; Drapeau *et al.*, 2009; Kumfor *et al.*, 2014).

Emotionally salient stimuli may be linked to basic biological drives and are broadly relevant to social signalling, self-awareness and reinforcement learning in a number of dementia syndromes (Sturm *et al.*, 2006; Kumfor *et al.*, 2011; Chiong *et al.*, 2013; Sturm *et al.*, 2013; Perry *et al.*, 2014; Shany-Ur *et al.*, 2014). The networks involved in normal emotion processing involve regions affected in FTD (Seeley *et al.*, 2007a; Seeley *et al.*, 2007b; Kober *et al.*, 2008; Omar *et al.*, 2011b; Hsieh *et al.*, 2012b; Bertoux *et al.*, 2014). However, while emotional disturbances are inherent to these dementias, and potentially relevant to disease detection, tracking and therapy, the pathophysiology of disturbed emotion in dementia is poorly understood and challenging to measure objectively.

Hypotheses and Predictions: Given the overlap of the anatomical substrate of these processes with those involved in salience processing, I hypothesised that: I hypothesised that with greatest damage to fronto-insular regions, key to autonomic responses, bvFTD would show overall depressed pupillary responses to auditory stimuli, irrespective of valence, but that PNFA and AD, with more posterior disease burden, would show normal overall responses. I further predicted that the syndromes of bvFTD and SD, showing greatest emotional recognition deficits,

would display the greatest impairment in physiological reactivity as modulated by emotional valence and that the normal relationship between pupillary response and valence of sound would be preserved in both PNFA and AD. Further, I predicted that in SD deranged responses would potentially reflect a damaged semantic system rather than impaired core autonomic reactivity.

5.3 Methods:

5.3.1 Participant Characteristics:

Forty-six patients (14 bvFTD, 12 PNFA, 10 SD, 10 amnestic AD and 26 healthy age-matched individuals) participated in this study. Ten patients with bvFTD had a genetic diagnosis (five pathogenic C9orf72 mutations, five MAPT mutations).

General demographic and neuropsychological data for participant groups are summarised in Table 5.1. The experimental groups were well matched for age; males were significantly over-represented in the bvFTD group. Mean symptom duration was longer in the bvFTD group than other patient groups, reflecting the wide variation in disease tempo of patients with bvFTD; the syndromic groups were otherwise similar in overall disease stage. Average MMSE score was lower in the SD and AD groups than the healthy control group, but did not differ between patient groups. At the time of testing, four patients were receiving anti-depressant medication (1 AD, 1 PNFA, 2 SD), but were currently not displaying any symptoms of depression according the carers accompanying the patients to the research appointment.

5.3.2 Experimental stimuli and procedures:

5.3.2.1 Sound stimuli: In a pilot experiment, the sound stimuli generated for the semantic matching task described in chapter 2 were presented to a cohort of 20 healthy young adults and were rated for both affective valence and identifiability. Sound pairs where both sounds in the pair were not matched for valence (less than one valence point difference on a scale of 1-10

between the two sounds) were discarded to avoid any potential confounds of effects of valence on identifiability); this yielded a final stimulus set of 45 sound pairs used in a semantic matching task for the final experiment (Table 5.2).

For the pupillary experiment, a subset of sounds representing three emotional valence categories: 'unpleasant' (e.g., a person spitting, a mosquito), 'neutral' (e.g., telephone, throat clearing) and 'pleasant' (e.g., baby laughing, stream burbling) were selected. Sound valence categories had similar overall identifiability ratings and sounds in each valence category were matched for other psychoacoustic properties. Final stimulus characteristics are described in Tables 5.3. During the experiments, all sound stimuli were presented via headphones from a notebook computer at a constant, comfortable listening level (at least 70 dB) in a quiet room.

Characteristic	**Healthy controls**	**bvFTD**	**SD**	**PNFA**	**AD**	**ANOVA**
demographics						
No. in group	26	14	10	12	10	
Handedness (R:L)	25:01:00	13:01	08:02	11:01	10:00	
Gender distribution (m:f)	12:14	11:03	06:04	03:09	05:05	
Age (yrs): mean (range)	67 (57-74)	66 (52-84)	65 (56-78)	68 (57-79)	66 (60-78)	
Education score	17 (2)	15 (3)	15 (3)	15 (3)	15 (2)	
Symptom duration (yrs)	NA	8.8 (6) [b,c,d]	5.2 (2)	4.8 (2)	5.3 (2)	
No. receiving AchEI	NA	6	1	2	9	
neuropsychology						
MMSE (range)	30 (29-30)	25 (18-30)	**21 (9-29)**	28 (27-29)	**25 (21-29)**	$p<0.001$, $F=7.96$, $r^2=0.43$
Verbal	123 (8)	**89 (20)**	**80 (18)**[d]	**77 (15)**[d]	**101 (14)**	$p<0.0001$, $F=28.59$, $r^2=0.66$
Performance	119 (14)	**97 (17)**	110 (17)	**98 (17)**	**89 (16)**	$p<0.0001$, $F=8.91$, $r^2=0.38$
RMT words (/50)	47 (3)	**35 (6)**	**32 (7)**	40 (8)	**30 (5)**[a,c]	$p<0.0001$, $F=28.6$, $r^2=0.67$
RMT faces (/50)	44 (4)	**34 (6)**	**38 (8)**	**38 (5)**	**32 (5)**	$p<0.0001$, $F=9.57$, $r^2=0.39$
Stroop word	21 (4)	**27 (9)**	27 (9)	**50 (14)**[a,b,d]	**31 (9)**	$p<0.01$, $F=5.41$, $r^2=0.36$
Stroop inhibition	57 (16)	**94 (42)**	77 (32)	**118 (51)**	**116 (47)**	$p<0.0001$, $F=15.8$, $r^2=0.65$
Digit span reverse (max)	5 (1)	5 (1)	6 (2)	**3 (1)**[b]	5 (2)	$p<0.001$, $F=5.3$, $r^2=0.32$
Spatial span reverse (max)	7.6 (2)	**5.6 (2)**	**5.6 (2)**	**4.7 (1)**	**7.9 (2)**	$p<0.01$, $F=3.85$, $r^2=0.27$
VOSP (/20)	18 (2)	17 (2)	16 (3)	16 (2)	**16 (2)**	$p<0.0001$, $F=28.59$, $r^2=0.66$

Verbal semantics: BPVS (/150)	148 (2)	**132 (15)**	**99 (45)**[a,d]	**132 (24)**	**140 (8)**	p<0.0001, F=13.43, r^2=0.49
SMT score **(45)	40 (5.2)	**35 (10.9)**	**35 (8.1)**	38 (6.2)	38 (7.1)	p=0.001, F=5.65, r^2=0.36

Table 5.1. Demographic, clinical and neuropsychological characteristics of participant groups Maximum total scores are shown (where applicable) after relevant neuropsychological tests; mean (standard deviation) data are shown unless otherwise indicated. Initial one-way ANOVA results between groups where statistically significant group differences ($p<0.05$) are shown in the right column and significant group deficits ($p<0.05$) versus the healthy older control group are shown in bold. Other significant differences ($p<0.05$) between groups are indicated by superscripts, a, relative to; a, bvFTD; b, SD; c, PNFA; and d, AD groups.

'same' sound pairs		valence 1	valence 2	mean valence
woman crying	woman screaming	1.8	1.3	1.6
man shouting in pain	man vomiting	2.1	1.4	1.7
car crash sounds	car horn	1.9	1.9	1.9
car horn	car skidding	1.9	2.2	2.1
man sobbing	person breaking wind	2.3	2.5	2.4
infant wailing	infant sobbing	2.6	2.3	2.4
person coughing	person snoring	3.6	2.8	3.2
woman clearing throat	woman yawning	4.6	4.9	4.7
geese honking	goose wings flapping	5.1	4.2	4.6
grandfather clock chime	grandfather clock ticking	6.5	5.6	4.6
train travelling on tracks	train horn	5.5	4.7	5.1
cockerel crowing	hen clucking	5.9	5.7	5.8
pigeon wings flapping	pigeon cooing	5.7	6.3	6
woman giggling	woman humming	7.4	7	7.2
stream burbling	waves lapping	7.7	8.4	8.1
baby cooing	baby laughing	7.8	8.8	8.3
Condition mean valence				*4.4*
'different' sound pairs		valence 1	valence 2	mean valence
mosquito	woman screaming	1.4	1.3	1.3
man vomiting	clock alarm bell	1.4	2.2	1.8
woman crying	car crash sounds	1.8	1.9	1.8
man sobbing	bees humming	1.8	1.9	1.9
telephone receiver replace	woman coughing	1.8	2.1	1.9
infant wailing	man wheezing	2.6	2.1	2.3
man shouting in pain	car horn	2.1	2.9	2.5
car skidding	man snoring	2.2	2.8	2.5
car alarm disarmed	shovel on metal	3.3	4.2	3.7
telephone dial tone	man clearing throat	3.4	4.1	3.7
cat whining	puppy yelping	4.1	3.6	3.8
paper tearing	child hiccoughing	3.9	4.2	4
telephone ringing	person breathing	4.8	4.2	4.5
paper rustling	woman clearing throat	4.7	4.6	4.6
infant sneezing	engine running	4.5	4.8	4.6
person brushing teeth	train horn	5	4.7	4.8
shovel digging gravel	car window winder	5.3	4.7	5
person clapping hands	dog barking	5.3	5.1	5.2
horse whinnying	woman yawning	5.5	4.9	5.2
child yawning	geese honking	5.7	5.1	5.4
pigeon wings flapping	person chewing	5.7	5.3	5.5
hen clucking	man sighing	5.7	5.3	5.5
fizzy drink can opened	coin dropped on table	6.2	6.2	6.2
pigeon cooing	fingers clicking	6.3	6.2	6.3
horse trotting	person walking on gravel	6.8	6	6.4
woman humming	water trickling	7	6.6	6.8
waves crashing	woman giggling	7.6	7.4	7.5
baby cooing	stream burbling	7.8	7.7	7.8
baby laughing	waves lapping	8.8	8.4	8.6

Table 5.2. Experimental stimuli for the auditory semantic classification experiment: pairs in 'same' and 'different' conditions, here ordered by mean affective valence assigned each sound pair; sounds in each pair were closely matched for valence (within 1 rating point). Valence ratings are derived from the pilot healthy younger control group (see Table 5.3). Sound pairs were presented in randomised condition order during the experiment

Trial no.	Sound name	Valence categ	Young control	Older controls			bvFTD		SD		PNFA		AD	
			mean valence	ident	mean valence	s.d.	mean valence	s.d.	mean valence	s.d	mean valence	s.d.	mean valence	s.d.
1	infant wailing	1	2.6	100	2.9	1.5	4.6	3.1	4.1	2.5	3.1	2.2	5.0	2
2	paper rustling	2	4.7	90	4.4	1.4	4.2	2	3.4	2.4	4.3	2.4	4	1.7
3	baby cooing	3	7.8	100	6.7	1.4	5.4	2.7	6.6	2.5	5.6	2.9	6.6	1.7
4	water trickling	3	6.6	100	6.1	1.5	6.3	2.2	6.4	1.8	5.2	2.3	5.4	1.8
5	bees humming	1	1.9	90	2.8	1.2	3.4	2.2	4.4	2.8	2.3	0.9	3.3	0.8
6	woman yawning	2	4.9	100	5.6	0.9	6.1	2.9	6.5	2.3	5.1	2.6	5	1.9
7	person spitting	1	1.7	90	2.5	1.4	3.2	1.4	3.5	2.8	2.4	1.3	2.3	1.1
8	child hiccoughing	2	4.2	90	3.3	1.1	3.4	1.9	4.3	3.1	3.4	1.9	3.4	1.5
9	fizzy drink poured	3	7.6	100	6.4	1.3	5.9	2.3	5.8	1.9	6.1	2.6	5.4	2.1
10	thunder	2	5.2	100	5.1	2.2	2.8	2.0	4	2.5	4.5	2.4	3.6	1.6
11	waves lapping	3	8.4	100	5.8	1.9	6	2.8	4.8	2.4	6.7	1.9	3.5	1.9
12	telephone receiver replace	1	1.8	90	3.6	1.2	2.5	1.1	4.1	1.6	3.4	2.4	3.7	1.7
13	man belching	1	1.6	90	2.6	1	2.4	1.6	3.8	2.6	1.9	0.7	2.2	1.2
14	woman giggling	3	7.4	100	6	1.7	7	2.2	6	2.5	6.9	1.9	4.8	1.8
15	train horn	2	4.7	90	5.8	1.3	5.2	2.7	5.6	2.7	5.5	2.7	3.7	1.6
16	woman humming	3	7	100	6.9	1.2	7.5	1.6	5.7	3.8	7.1	1.4	6.6	2.7
17	car horn	1	1.9	100	3.4	1.1	3.5	1.9	4.3	2.7	3	2	2.3	0.6
18	woman clearing throat	2	4.6	100	4.7	0.7	4.8	2.1	5.1	2.3	3.1	1.6	4.5	1.6
19	man vomiting	1	1.4	100	1.8	0.8	1.8	0.5	2.7	2.3	1.8	0.6	1.7	0.7
20	telephone ringing	2	4.8	90	4.4	0.9	3.1	1.6	5.7	2.5	5.5	1.9	3.8	1.2
21	stream burbling	3	7.7	100	6.4	1.6	6.1	2.7	6.1	2.2	6.5	2.4	4.6	1.1
22	infant sneezing	2	4.5	90	3.2	1.3	2.9	1.3	2.9	1.3	2.9	1.1	2.7	1
23	woman crying	1	1.8	100	3	0.9	2.9	1.4	2.8	1.3	2.4	1.4	2.5	1.5
24	waves crashing	3	7.6	100	5.8	1.6	5.2	3.0	3.6	1.6	5.4	2.5	4.3	2.8
25	horse trotting	3	6.8	100	6.4	1.2	6.2	1.8	5.3	3.1	7.2	1.6	5.8	1.9
26	person brushing teeth	2	5	100	4.6	0.9	4.9	2.3	4.8	2	5.3	2.6	5.1	1.5
27	mosquito	1	1.4	100	2.6	1.2	3.7	2.2	4.5	3.1	2.5	1.9	3.2	1.5
28	woman screaming	1	1.3	100	2.3	0.9	2.1	0.8	2.8	1.5	2.4	2	1.6	0.6
29	car engine idling	2	4.8	100	5.1	1.2	5	2.3	5.3	2.2	4.5	2.6	4.2	1.8
30	baby laughing	3	8.8	100	7.1	1.1	7.6	1.8	7	2.3	7.6	1.7	6.6	2.2

Table 5.3. Experimental playlist and sound stimulus psychological characteristics for the pupillometry experiment. Sound stimuli are listed in trial presentation order. The healthy younger control group (n=20, median age 28 years (range 23-37), 6 male) participated in an initial pilot experiment on a larger set of 180 nonverbal sounds that were rated for affective valence (pleasantness, 1 - 10) on a Likert scale. For participant groups in the main experiment, affective valence ratings (mean and standard deviation, s.d.) assigned to each individual sound by the relevant group are shown. **categ**, valence category: 1= negative, 2= neutral, 3= positive; **ident**, for each of the 30 sounds, the proportion of the healthy older control group correctly identifying that sound is shown. Mean proportion of sounds correctly identified was similar between valence categories (valence category 1, 96%; category 2, 96% ; category 3, 100%). The first three practice trials are shaded in grey.

5.3.2.2 Pupillometry:

Pupil dilatation responses were measured for 27 sounds (nine from each valence category), presented in randomised order (see Table 5.3); three additional sounds were presented as an initial familiarisation set but not further analysed. On completion of pupil recording for each trial, a modified Likert scale was displayed and the participant was asked to rate the pleasantness (affective valence) of the sound. All pupil response and behavioural rating data were stored for off-line analysis.

5.3.2.3 Data analysis:

Pupil response and behavioural affective valence rating data were compared between participant groups in three ways. Firstly, as the key aims of this experiment were to provide novel biomarkers for stratifying individual differences in autonomic responses or behavioural interpretation of hedonic stimuli, an individual's pupillary responses were compared to their own behavioural rating using a linear mixed effects model with crossed random effects for participant and sound. Secondly, to investigate deviation from the normal autonomic response to a sound of a given valence, again using a linear mixed effects model with crossed random effects for participant and sound, an individual's pupillary response for each sound was correlated with the 'normal' valence of that sound (i.e. the averaged response of healthy older controls). Thirdly, as there was considerable noise in the data, within each group, averaged pupillary responses in relation to averaged valence rating for each sound were analysed using linear regression (as there was one data point per sound a linear mixed effects model was no longer necessary). In all analysis the relationship between pupillary responses and sound valence were compared directly and quadratically $(\text{sound valence})^2$ in order to capture any quadratic association with pupil response (since pupil response was anticipated to increase both for highly positively- and negatively-valenced sounds). Coefficients of the quadratic relationship between pupil response and valence (i.e. a measure of the strength of the curvlinear relationship) were additionally compared between groups.

Variability within each group of individual Pupilmax responses and affective valence ratings was assessed by calculating the difference between an individual's rating or Pupilmax response and the mean for that group and linear regression models were used to compare participant groups.

Measures of correlation strength (r^2 values) between pupil response and affective valence were generated for each group. Clinical symptom duration, MMSE score and reverse spatial span (a cognitive measure of nonverbal executive function and working memory) were taken as surrogates of disease severity across syndromes and correlations of these disease measures, peripheral hearing function and both anti-depressant and Acetyl-choline Esterase inhibitor medication use with pupil reactivity and auditory affective valence ratings were assessed in the patient cohort. Relations between auditory affective ratings and performance on the nonverbal auditory semantic test were separately assessed.

5.4 Results:

5.4.1.1 Behavioural affective valence rating profiles:

Mean affective valence ratings assigned to each sound by the healthy older control group and the healthy young pilot control group were strongly positively correlated (r^2=0.96, p<0.0001); valence ratings assigned to the sound stimuli by all groups are listed in Table 5.3.

Mean auditory affective valence ratings of each patient group relative to the healthy older control group are plotted in Figure 5.1. Across the sound stimulus set, mean valence ratings for each patient group showed a significant positive correlation with control mean ratings (bvFTD vs. control, p<0.0001, F=127.62, r^2=0.84, t=11.3; SD vs. control, p<0.0001, F=; 45.9, r^2=0.65, t=6.77; PNFA vs. control, p<0.0001, F=230.13, r^2=0.9, t=15.17; AD vs. control, p<0.0001, F=91, r^2=0.78, t=9.54). The AD group rated sounds overall as significantly (p=0.02, z=-2.39, $\mathcal{X}^2$=11.51) less pleasant than the other groups; there were no other group differences

for overall valence profile, though particular sounds were rated as less pleasant by each of the patient groups relative to the healthy older control group (Figure 5.1).

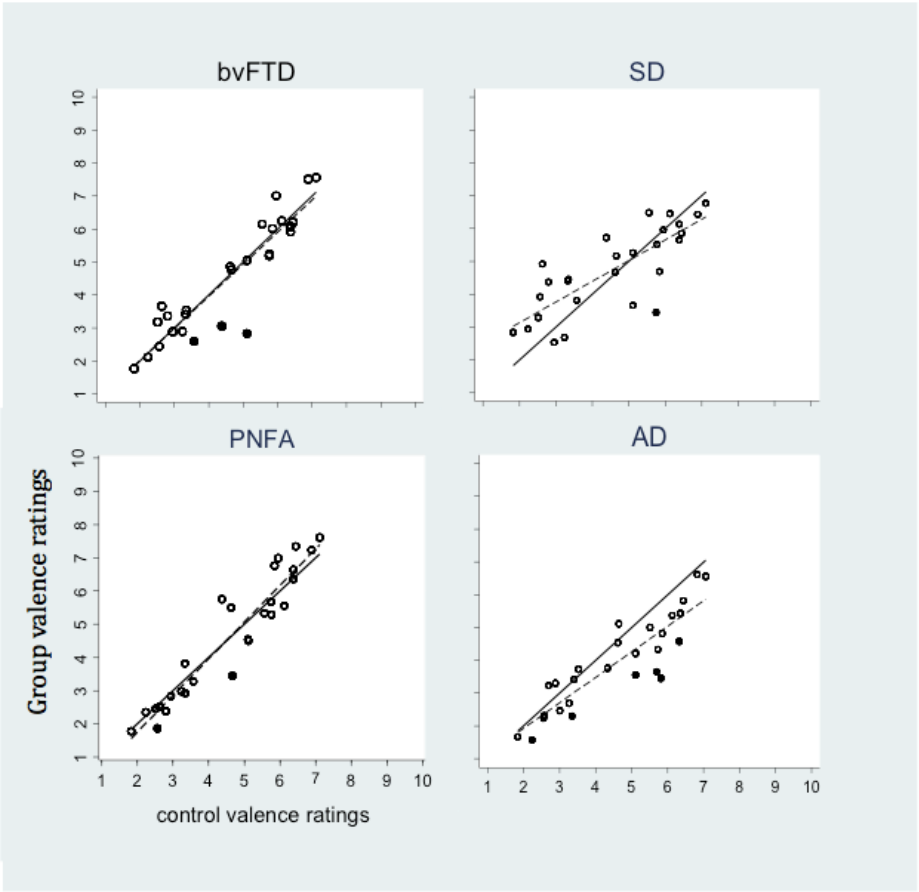

Figure 5.1. Mean group affective valence (pleasantness) rating for each stimulus sound plotted against healthy older control group mean affective valence ratings, for each patient group. Ratings are on a Likert scale where 1 and 10 indicate most unpleasant and most pleasant, respectively. For ease of visualisation, lines of best fit for control group ratings (solid line, x=y) and patient group ratings (dashed line) are plotted. Black filled squares code particular sounds for which mean valence ratings were significantly different between patients and healthy older controls.

5.4.1.2 Pupillometric data:

For all participant groups, pupil dilatation began around 0.25 s after sound onset and peaked around 1.25 s (Figure 5.2). Baseline pupil size did not differ significantly between groups; the bvFTD, SD and AD groups showed a reduction of baseline pupil size but not Pupilmax over the course of the experiment. Mean Pupilmax values over the entire sound stimulus set (indexing overall pupil reactivity to sound) were normal in the AD group but significantly reduced relative to both healthy controls and the AD group in the other patient groups (controls vs. bvFTD, $p<0.05$, $F=12.44$, $r2= 0.28$, $t=--1.99$; controls vs. SD, $p<0.0001$, $F=12.44$, $r2= 0.28$, $t=-6.03$; controls vs, PNFA, $p=0.001$, $F=12.44$, $r2= 0.28$, $t=-3.52$; AD vs. bvFTD, $p<0.05$, $F=11.41$, $r2= 0.26$, $t=-1.67$; AD vs. SD, $p<0.0001$, $F=15.65$, $r2=0.29$, $t=5.58$); the SD group showed a smaller mean overall Pupilmax response than all other groups and correspondingly smaller overall individual variability in pupil responses (SD vs. bvFTD, $p<0.0001$, $F=11.41$, $r2=0.26$, $t=-3.96$; SD vs. PNFA, $p=0.2$, $F=15.65$, $r2=0.29$, $t=2.43$).

Within the healthy control group, examining the quadratic relationship between pupillary response and valence of the presented sound for each individual *separately* did not yield consistent results; whilst some individuals demonstrated a clear curvelinear relation between the valence of the sound and the magnitude of the pupillary response, in other individuals no clear correlation could be shown. Similarly, comparing trends within a group between an individual's pupillary response and the valence of a sound did not yield clear correlations. Additionally an initial one-way ANOVA to assess for group differences in the coefficients of the quadratic relationship between pupil response and valence (i.e. a measure of the strength of the curvlinear relationship) did not show any significant result.

However, when each individual's pupillary response to each sound was then compared to the *average* valence of the sound for the 'normal' valence (average of the control group's response), there was a significant curvlinear relationship between each individual's response

and the normal valence of the sound in both the healthy older adults ($p<0.01$, $z=3.34$, $\chi^2=11.57$), and the PNFA group ($p=0.04$, $z=1.95$, $\chi^2=6.18$).

When comparing the individual's pupil response for each sound to the valence for that sound averaged for the group to which they belong, this correlation remained in the PNFA group ($p=0.02$, $z=2.47$, $\chi^2=7.49$) and a correlation emerged in the SD group ($p=0.04$, $z=2.12$, $\chi^2=6.23$).

Comparing averaged pupil response to a given sound to the averaged affective rating for that sound within each group, the healthy older control group showed a significant curvilinear relation ($r^2=0.44$, $p<0.01$, $t=3.21$, $F=5.91$) between Pupilmax and affective valence ratings, with significantly greater pupil responses to both highly pleasant and unpleasant sounds than to neutral sounds (Figure 5.3). When referenced to the affective valence ratings for the corresponding patient group, both the PNFA group ($p<0.01$, $F=3.98$, $r^2=0.34$, $t=3.10$) and the SD group ($p<0.01$, $F=3.42$, $r^2=0.31$, $t=2.93$) but not the other patient groups showed significantly increased pupil responses to highly valenced sounds (Figure 5.3). This correlation was lost in the SD group if pupil responses were referenced to healthy control (rather than patients' own) valence ratings. Coefficients of the relation between Pupilmax and affective valence did not differ significantly between groups. Plotting pupillary responses against normal sound valence revealed wide individual variability of pupil responses across the sound stimulus set in all participant groups (Figure 5.4). Pupillometric and behavioural valence rating profiles of syndromic groups relative to healthy older controls are summarised in Table 5.4.

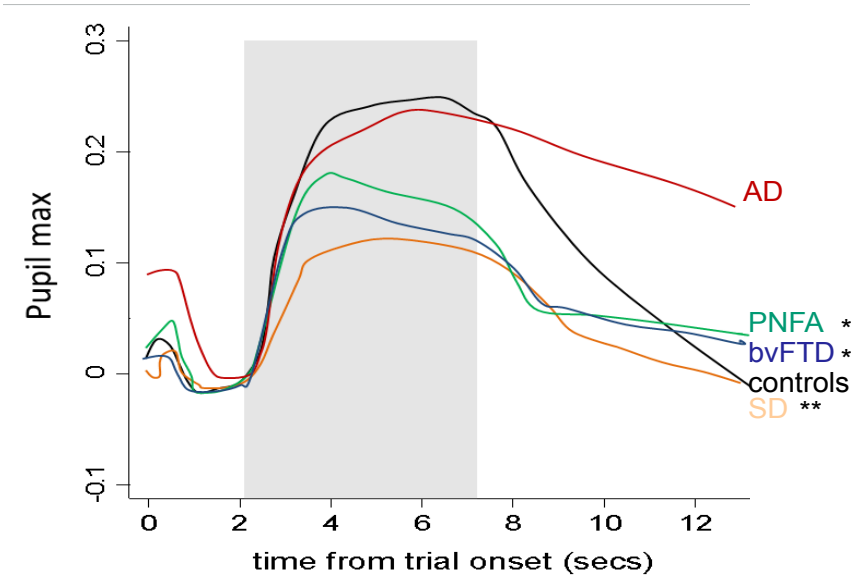

Figure 5.2. The mean time course of pupil response, Pupilmax over all trials is plotted for each participant group. The shaded grey time block indicates sound stimulus presentation. Mean pupil responses were normal in the AD group, reduced in the bvFTD and PNFA groups relative to the control and AD group (*) and reduced in the SD group relative to all other groups (**).

5.4.1.3 Associations with general disease measures and auditory semantic function:

There was no evidence that affective valence ratings, overall pupil reactivity or pupil responses to sound valence correlated with disease severity (as indexed by nonverbal executive impairment, MMSE score or symptom duration), peripheral hearing function or medication use.

The healthy older control group achieved sub-ceiling scores on the sound pair semantic classification task; relative to controls, the PNFA and AD groups showed no auditory semantic deficit while both the SD and bvFTD groups showed significantly impaired performance (controls vs. bvFTD, $p<0.001$, $F=10.52$, $r2=0.42$, $t=-5.10$; controls vs. SD, $p<0.0001$, $F=10.52$, $r2=0.42$, $t=-5.4$), and the SD group performed significantly worse than the PNFA group ($p<0.01$, $F=6.21$, $r2=0.35$, $t=3.24$) (see Table 5.1).

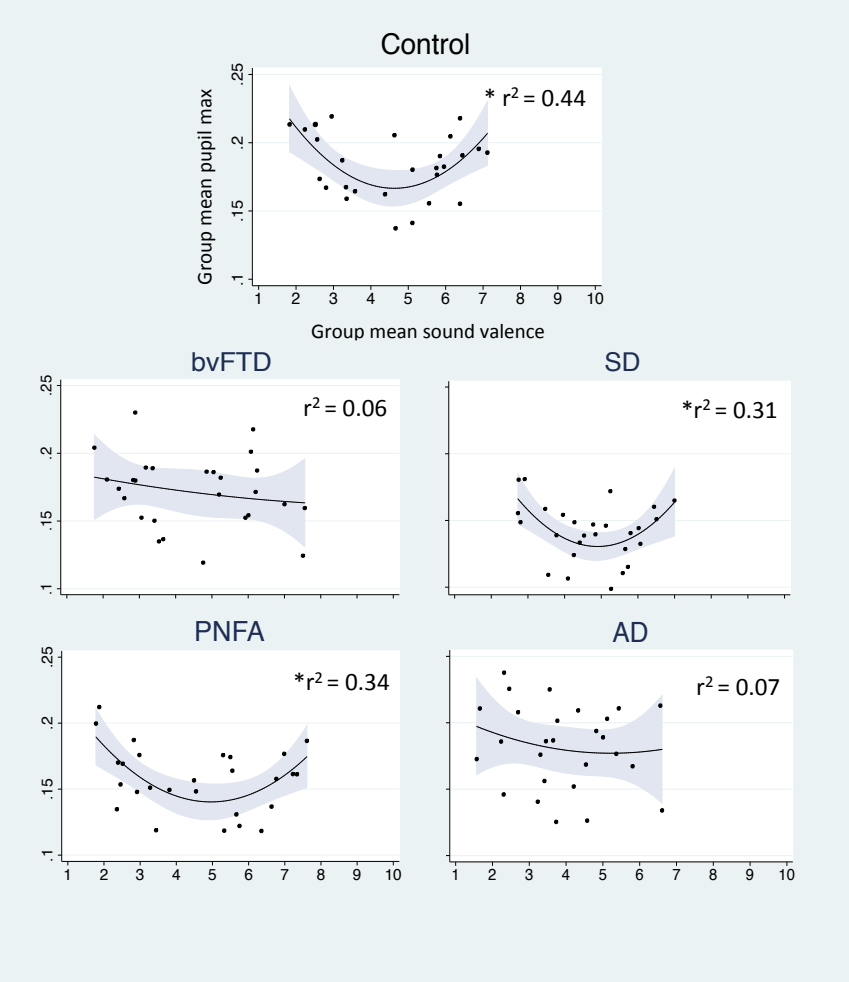

Figure 5.3. Pupilmax in response to each stimulus sound plotted against own group mean affective valence (pleasantness) ratings, for each participant group. Quadratic regression lines of best fit with 95% confidence intervals (shaded grey zones) and corresponding r^2 values are shown. *significant ($p<0.05$) correlations between pupil response and sound valence

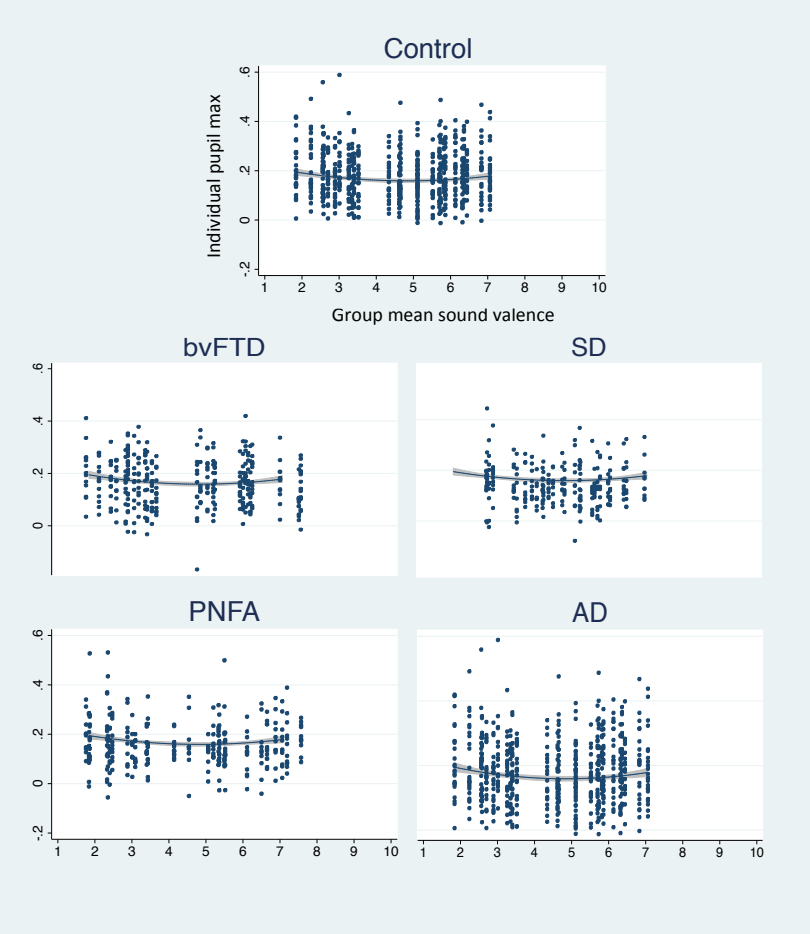

Figure 5.4 Individual Pupilmax in response to each stimulus sound plotted against group mean affective valence ratings, for each participant group. Quadratic regression lines of best fit with 95% confidence intervals (shaded grey zones) are shown.

Disease group	Pupil responses		Valence rating	Semantic performance**
	Overall reactivity	Valence coupling*		
bvFTD	Impaired†	Impaired	Preserved	Impaired
SD	Impaired††	Preserved¶	Preserved	Impaired§
PNFA	Impaired†	Preserved	Preserved	Preserved
AD	Preserved	Impaired	Impaired††	Preserved

Table 5.4. Summary of syndromic profiles of emotional sound processing relative to healthy controls. *correlation of pupil response with affective sound valence ratings by that group; **nonverbal auditory semantic classification task; †also relative to AD group; ††relative to all other groups; ¶impaired if referenced to healthy control (rather than patients' own) affective ratings; §also relative to PNFA group

5.5 Discussion:

Here I have shown that, relative to healthy older individuals, patients with dementia syndromes have distinctive and partly dissociable profiles of autonomic (pupillary), behavioural and cognitive responses to emotionally salient nonverbal sounds (Table 5.4).

With regards to the first hypothesis that bvFTD would show overall depressed autonomic responses to auditory stimuli, irrespective of valence, but that overall reactivity would be preserved in PNFA and AD, patients with AD showed retained overall pupillary responses to sound whist overall reactivity was depressed in the bvFTD and SD FTD subtypes, consistent with previous studies (Robles Bayon, 2000; Sturm *et al.*, 2008), but also in PNFA. This may reflect limitations of sampling size when applied to a heterogeneous group (See chapter 8 for further discussion). Although this study was not set up for imaging analysis, impairment of overall reactivity in FTD is in keeping with the involvement of areas central to autonomic response generation in these diseases; in particular the anterior insula (Harper *et al.*, 1998; Critchley *et al.*, 2000a; Critchley, 2005; Menon & Uddin, 2010; Hsieh *et al.*, 2012a; Beissner *et al.*, 2013) and is consistent with previous work demonstrating depressed autonomic reactivity in FTD (Sturm *et al.*, 2008; Robles Bayon *et al.*, 2014). Anatomical substrates overlap by syndrome, however in bvFTD depressed responses may be more driven by anterior insula mediated

general salience dysfunction (Zhou and Seeley, 2014) and in SD an additional component of further amygdala damage may play a part in their additionally significantly depressed responses relative to the other FTD subtypes (Whitwell *et al.*, 2005). That overall pupillary reactivity can differentiate between bvFTD and AD may have the potential to help stratify these diseases on a physiological level. Whilst within this study the differences in overall pupillary responses were best demonstrated at group level, and one must therefore remain reticent about overstating interpretation of differences on an individual level, this does suggest that an evolved form of these measures (perhaps by combining with other autonomic metrics) may have a role in the clinical or research setting in differentiating those cases where clinical phenotype is ambiguous. Further, extending this work to investigate pre-symptomatic known gene mutation carriers, especially in conjunction with longitudinal measurements, may reveal physiological changes before clinical ones become apparent, changes that can map progression of disease with greater sensitivity than currently used metrics.

With regards to the hypothesis, that normal autonomic responses to emotional stimuli would be lost in bvFTD and SD relative to PNFA and AD, the results are less clear. That normal effects of sound valence upon autonomic responses should be effectively retained in PNFA is consistent with the fewer emotional deficits observed clinically and the lesser involvement of areas key to emotional processing (Kumfor & Piguet, 2012). However, in AD as well as bvFTD and SD, this relationship was lost. This held true both against the normal valences of a sounds (as rated by the control group) and the valence as rated by the relevant disease group, and therefore deficits could lie in either or both affecter (emotional interpretation) and effector (autonomic response generation) pathways. In bvFTD this is consistent with previous work (Sturm *et al.*, 2006; Werner *et al.*, 2007; Sturm *et al.*, 2008; Femminella *et al.*, 2014; Robles Bayon *et al.*, 2014). However, the result in the AD group is harder to explain; if they indeed have heightened salience processing, one might have anticipated an exaggeration of the normal curvelinear relationship one observes between autonomic response and valence in healthy

older controls. Alternatively, a generally disrupted but 'heightened' salience processing system may render all sounds (irrespective of valence) more salient in AD (which would be consistent with their behavioural ratings of the pleasantness of sounds being overall skewed towards being more unpleasant). In SD coupling of autonomic with behavioural responses as indexed by patients' own valence ratings was retained, however, this coupling was lost if referenced to healthy control ratings. This is consistent with the initial hypothesis that suggests that patients with SD retain the ability to cognitively interpret emotional stimuli, and mount appropriate physiological responses as modulated by sound valence, but have a distortion of which stimuli are emotionally evocative to them. One would hypothesise, in light of their worse auditory semantic performance that this reflects a mis-interpretation of environmental sounds.

Certain limitations and caveats to this work must be discussed: Firstly, within the healthy control group examination of the relationship between pupillary response and valence of the presented sound for each individual separately did not yield consistent results, with some individuals demonstrating a clear curvelinear relation between the valence of the sound and the magnitude of the pupillary response, whilst in other individuals no clear correlation could be demonstrated. This likely reflects the generally 'noisy' nature of pupillary measurements and highlights an inherent limitation of the stimuli set used; in order to develop specific biomarkers that could be used on an individual level, a consistent response would need to be demonstrated initially in the healthy control group before abnormality could be shown in disease groups. Secondly, when a random effects model has been applied, this has been by sound rather than by individual which limits the extrapolation to other patients beyond those in the study. Thirdly, individual variation in pupil responses and affective valence ratings was substantial and heightened in the patient cohort compared with healthy older individuals. Additionally, while affective rating profiles of the bvFTD, SD and PNFA groups were similar overall to the healthy control group, particular sounds elicited discrepant valence ratings in these patient groups. It

remains unclear whether this is simply a sampling issue or whether these sounds might tap more subtle disease-associated alterations in emotional salience coding.

Further studies should aim to use larger group sizes; the validity of the autonomic and behavioural metrics we have identified should be assessed in larger cohorts incorporating defined molecular pathologies and longitudinally, in order to define the time course of physiological alterations over the evolution of these diseases, including presymptomatic carriers of pathogenic mutations. Additionally, at the time of testing 4 patients were on anti-depressant medication (1 AD, 1 PNFA, and 2 SDs). Potentially depression could have affected the magnitude of the pupillary responses as depression is known to have an effect on emotional reactivity (Dixon-Gordon et al., 2015). Given the small number of patients with on anti-depressant medication and the lack of clinical signs of depression as reported by the patients' carers at the time of testing, I feel this is unlikely to have had an impact upon the results, especially when comparing group differences. However, further work should factor in depressive rating scales (for example the HADs scale) to assess for any impact of mood upon physiological response. The neuroanatomical correlates of the autonomic and behavioural metrics identified here remain to be defined: functional neuroimaging paradigms, ideally incorporating dynamic techniques such as magnetoencephalography with direct autonomic correlation will enable direct evaluation of candidate brain mechanisms. Ultimately, pathological correlation including detailed histomorphometry of key components of central autonomic circuitry will be required to establish the sensitivity and specificity of physiological markers for particular tissue pathologies and to define their brain substrates directly. Emotional sounds and pupillometry measures should be assessed alongside alternative stimulus paradigms and autonomic effector modalities tailored for particular behavioural signatures and diseases, and specific components of the behavioural affective response (in particular, valence and arousal) should be differentiated (Gray *et al.*, 2009; Beissner *et al.*, 2013; Zhou & Seeley, 2014). Autonomic indices will need to be correlated with clinical symptoms and disability to assess their functional relevance. Potential modulating effects of autonomically

active drug classes should also be assessed, in order to interpret clinical data in patients receiving these agents, and further, to test specific pathophysiological hypotheses (concerning, for example, aberrant reinforcement learning: (Perry *et al.*, 2014)) and to dissect the relative contributions of sympathetic and parasympathetic control mechanisms.

5.6 Chapter conclusions:

Here I have demonstrated that autonomic reactivity to sound is depressed in FTD relative to AD and controls, and with a larger sample size, in SD and bvFTD also relative to PNFA, in keeping with impaired circuitry involving anterior aspects of the salience circuitry. With regards to the modulation of autonomic responses by emotional content of the stimuli, in PNFA, autonomic emotional reactivity remained normal, whilst patients with bvFTD, SD and AD showed abnormal coupling between pupillary and affective behavioural responses to emotionally salient sounds. However, in SD normal autonomic responses can be mounted, but emotional interpretation is impaired, likely reflecting disrupted underlying semantic mechanisms. Different autonomic profiles to affective sounds in dementias may help better define pathophysiological mechanisms and provide potential for the evolution of new physiological biomarkers.

Chapter 6: Salience of semantics

6.1 Chapter summary:

The semantic content of sounds may potentially act as a strong salience cue, which has significant biological implications. SD, as the canonical exemplar of a disrupted semantic system provides a window for investigating this further, and differentially affected signal processing relative to other FTD types may provide an avenue for physiologically differentiating diseases. Impaired sound identification was associated with increased pupil reactivity to real (meaningful) versus acoustically-matched synthetic (meaningless) sounds in healthy controls; this response was exaggerated in the SD group and could differentiate the SD group from the healthy older controls. Further, in all groups with an auditory sematic deficit, the magnitude of the differential response to the two sound conditions correlated with degree of semantic impairment. This physiological signature of altered auditory semantic salience had a neuroanatomical correlate in right anterior temporal pole. The results provide evidence that semantic content of environmental sounds are strong salience cues and may offer metrics for physiologically phenotyping dementias.

6.2 **Introduction:**

In order to interpret the continuous complex stream of sensory input from one's environment, an organism must be able to successfully disambiguate biologically relevant, 'salient' stimuli from the busy multisensory background such that attentional resources can be allocated appropriately. Some of this salience information is carried by low level perceptual cues, with pupil dilatation correlating with degree of stimulus contrast (Wang *et al.*, 2014) and perceived motion (see chapter 4), likely mediated by a salience processing system focused on

fronto-insular cortices (Seeley *et al.*, 2007b). However, on neurobiological grounds, it seems likely that the semantic content of a source stimulus will also provide markers of relative salience; think of the challenge of disambiguating and proportioning appropriate attention to two perceptually similar sounds with very different biological implications (compare, for example, the rumble of thunder and the growl of a large predator). Further, in the previous chapter we observed that patients with SD, despite abnormal autonomic reactions as modulated by the *normal* valence of environmental sounds, displayed a restoration of the valence-pupillary response curve when modulated by *their* perceived valence of sounds; a finding that may speak to distorted salience attribution based upon semantic misinterpretation of stimuli.

Hypotheses and predictions: The physiological and neuroanatomical correlates of this potential 'semantic salience' have yet to be established. Here I hypothesised that, due to their greater biological salience, usually meaningful sounds would evoke greater pupillary responses than acoustically matched meaningless counterparts and that an impaired semantic system would render these meaningful sounds less salient and result in a loss of this normal differential response. This would provide a physiological metric of semantic ability with the potential to differentiate and track disease progression. Patients with FTD and AD were presented with a series of both meaningful and meaningless sounds with pupil and behavioural responses measured.

6.3 Methods:

6.3.1 Participant characteristics:

Thirty-six patients (AD, n=10; bvFTD, n=10; SD, n=10; PNFA, n=6) and 20 healthy older individuals with no history of neurological or psychiatric illness participated in this study. Background demographics and general neuropsychometric data are given in table 6.1.

6.3.2 Experimental Design and methods:

6.3.2.1 pupillometry experiment design:

From the sound set used for determination of baseline non-verbal auditory semantic function outlined in chapter 2, ten environmental sounds rated as highly identifiable by 20 healthy younger individuals (median age 28 years (range 23-37), six male) were selected, see table 6.2 for the sounds and their mean valence and alerting ratings). White noise counterparts with matched frequency bandwidths and were generated in Matlab© and then spectrally shaped in Goldwave© ('meaningless', M-) as versions of the sounds that were acoustically similar but lacking the semantic associations of the real ('meaningful', M+) sounds. The M+ and M- sounds had matched mean overall sound intensity. Controls were able to identify all real sounds correctly and there was no concordance in estimating the sound origin of the synthetic sounds. Sounds in the M+ and M- conditions were all presented twice in a pseudo-randomised order, yielding a combined playlist of 40 trials. Four additional sounds (two M+, two M-) were presented prior to the playlist proper as familiarisation trials but were excluded from the final analysis.

Characteristic	Healthy controls	bvFTD	SD	PNFA*	AD*	ANOVA
General						
No.	20	12	11	6	10	
Gender (f:m)	10:10	10:02	07:04	01:05	05:05	
Age (y): mean (range)	65.6 (57-71)	65.2(52-76)	66.5 (53-78)	69.2(61-77)	69.0 (54-78)	
Education (y)	16.9 (12-20)	**14(11-21)[c]**	**14.7 (11-20)[c]**	18 (17-20)	15.2 (12-17)	$p<0.01$, $F=4.4$, $r^2=0.24$
Symptom duration (y)	NA	8 (3-21)	5.2 (3-9)	5.7 (4-10)	5.8 (3-8)	
IQ						
Verbal	125.1 (112-137)	**84.8 (55-116)[d]**	**80.7 (55-109)[d]**	**92.8 (70-115)**	**101.4 (81-129)**	$p<0.0001$, $F=22.32$, $r^2=0.62$
Performance	122.2 (99-141)	**98.2(70-135)**	**108.9 (88-135)**	**102 (83-121)**	**87.5 (66-112)[b]**	$p<0.0001$, $F=9.89$, $r^2=0.42$
Episodic memory						
RMT words (/50)	47.5 (42-50)	**32.9 (25-48)**	**35.9 (25-47)**	**37.4 (34-40)**	**29.4 (18-43)**	$p<0.0001$, $F=16.72$, $r^2=0.56$
RMT faces (/50)	43.35 (35-50)	**36.9 (25-48)**	**32.5 (25-45)c**	43.8 (41-46)	**33.1 (23-40)c**	$p<0.001$, $F=6.61$, $r^2=0.34$
Executive functions						
Stroop word	21.3 (15-30)	26.3 (18-39)	26.1 (14-38)	**52.8 (43-72)[a,b]**	NA	$p<0.0001$, $F=27.01$, $r^2=0.66$
Stroop inhibition	51.3 (35-70)	**99.5 (48-180)**	**80.6 (36-136)**	**121.6 (75-180)**	NA	$p<0.0001$, $F=1.06$, $r^2=0.44$
Digit span reverse (max)	5.5 (3-7)	4.5 (3-6)	5.6 (3-8)	4.2 (3-7)	5.3 (3-8)	
Visuoperceptual functions						
VOSP (/20)	18.9 (16-20)	**17.2 (13-20)**	**17.4 (14-20)**	18.2 (16-19)	**15.5 (12-18)[c]**	$p<0.01$, $F=5.16$, $r^2=0.29$
Semantic processing						
BPVS (/150)	148.4 (146-150)	**132.1 (102-147)**	**96.7 (41-147)[a,c,d]**	140.6 (131-145)	140.3 (120-148)	$p<0.0001$, $F=11.31$, $r^2=0.47$
Synonyms (50)	48.7 (48-50)	**36.8 (20-47)**	**34.0 (20-49)**	41.0 (31-48)	44.3 (41-46)	$p<0.001$, $F=6.44$, $r^2=0.42$
SMT score (60)	57.5 (51-60)	**48.2 (38-57)[c]**	**50.0 (40-57)[c]**	56.3 (54-59)	**51.0 (43-55)[c]**	$p<0.0001$, $F=13.04$, $r^2=0.5$

Table 6.1. Demographic, clinical and neuropsychological characteristics for experimental groups. Maximum total scores are shown (where applicable) after relevant neuropsychological tests; mean (range) data are shown unless otherwise indicated. Significant group differences versus the healthy older control group are shown in bold. Other significant differences between groups are indicated by superscripts: a, relative to bvFTD; b, SD; c, PNFA; and d, AD groups.

	control		bvFTD		SD		PNFA		AD	
Real sounds (M+)	**v**	**a**	**v**	**a**	**v**	**a**	**v**	**a**	**v**	**a**
Bees	3	7.2	3.3	7.2	3.1	6.8	3	6.1	3.4	7.2
Car horn	4	7.2	4.1	7.7	5.4	7.6	3.5	6	3.5	7.5
Motor engine	4.1	6.4	4	7.3	5	6.1	4.1	5.6	3.9	6.9
Phone being hung up	4	6.3	4.2	6.7	3.2	6.8	3.1	6	3.6	6.8
Waves lapping	4.4	6.7	4.1	7.3	4.3	7.3	3.7	6.3	4.7	7
Brushing teeth	4.4	6	3.9	7.6	4.7	6.5	3.6	5.8	4.5	6.2
Phone ringing	4	7.5	4.2	7.7	5.1	7.4	3.6	5.8	4.4	7.2
Babbling stream	5.5	6	5.4	6.3	5.2	6.3	4.9	5.5	5.5	5.7
Thunder	4.3	6.9	4.1	7.1	4.8	7.2	3.5	6.5	4.2	6.8
Train horn	4.3	7.5	4.6	6.9	5.5	6.9	3.8	6.3	4.4	6.7
mean	4.2	6.8	4.2	7.2	4.6	6.9	3.7	6	4.2	6.8
Synthetic sounds (M-)										
Bees	3.5	7.1	3.7	7.8	4.1	6.7	3.1	5.3	3.3	7.2
Car horn	4.1	6.8	4.1	6.8	3.9	6.5	3.2	6.3	4.2	6.6
Motor engine	3.8	6.8	3.8	7.1	4.6	7.3	3.2	6.1	4	7.1
Phone being hung up	3.4	7.1	3.9	7.4	4	6.8	3.3	5.4	3.3	7.4
Waves lapping	3.6	7	3.8	7.7	4.2	7.3	3	6.1	3.4	7.3
Brushing teeth	3.4	7.1	3.8	7	3.7	7.8	3.5	6.1	3.6	7.2
Phone ringing	3.5	6.9	3.6	7.1	4.3	6.5	2.9	6	3.8	7.2
Babbling stream	3.6	7	3.7	7.3	3.9	7.4	3.1	5.9	3.7	7.2
Thunder	3.8	6.8	4.1	7.2	3.9	7.3	3.4	6.1	3.9	6.9
Train horn	3.9	6.7	3.7	7.4	4	6.3	3.3	6.2	4.4	7.2
mean	3.7	6.9	3.8	7.3	4.1	7	3.2	5.9	3.8	7.1

Table 6.2. Real ('meaningful', M+) and synthetic counterparts generated from the real sounds ('meaningless', M-) stimuli presented in the pupillometry experiment with mean pleasantness (valence, v) and alerting (arousal, a) ratings in each group.

6.3.2.2 data analysis:

Statistical methods were used as outlined in chapter 2; the difference in mean pupil responses to the M+ and M- sound conditions was assessed within each group with paired t-tests. Differences in magnitude of response to the two conditions were compared with linear regression. In order to assess how well the M+ sounds presented in the pupillometry experiment indexed patients' general auditory semantic competence, I performed a separate sub-analysis of the semantic classification task described in chapter 2 to assess just those sounds used in this experiment (n = 14).

6.3.2.3 Brain image acquisition and analysis:

For 26 patients (12 bvFTD, 10 SD, four PNFA) a T1-weighted volumetric brain MR was available. Separate voxel-wise linear regression models were used to assess associations in the combined patient cohort between regional grey matter volume and parameters of interest: firstly, overall pupil reactivity (individual overall mean Pupilmax across the sound stimulus set) and secondly, differential pupil reactivity modulated by sound semantic content (individual difference in mean Pupilmax between the meaningful and meaningless sound conditions: M+ > M-). Anatomical small volumes encompassed the temporal lobes anterior to Heschl's gyrus (previously implicated in semantic analysis and signalling the behavioural value of sounds and other sensory objects: (Goll *et al.*, 2012), insular cortex (implicated as an 'autonomic hub' in salience processing in FTD: (Critchley *et al.*, 2000a; Seeley, 2010)) and dorsal brainstem including superior colliculi (previously identified as a key integrative site of autonomic effector response: (Wang *et al.*, 2012; Wang & Munoz, 2014)), yielding a total of 5 regions of interest.

6.4 Results:

Background neuropsychometric scores are shown for each group in table 6.1. Gender distribution and mean educational obtainment varied between participant groups and were therefore included as nuisance covariates in subsequent analyses of between group comparisons.

Baseline pupil size did not differ significantly between groups; the PNFA group showed a reduction of baseline pupil size over the course of the experiment, but Pupilmax showed no such trend in any group. Overall pupil reactivity for the combined sound stimulus set and for M+ and M- conditions separately did not differ between participant groups and did not correlate with patients' auditory semantic performance. M- sounds had higher peak loudness as measured with a hand held decibelometer and were rated as more unpleasant than (but equally alerting as) M+ sounds by the healthy control group and the effects of sound loudness and pleasantness were therefore adjusted for in the analysis. Pupil responses to M+ sounds were enhanced compared with M- sounds (Figure 6.1): this effect was a non-significant trend ($p=0.09$, $t=1.79$) in the healthy control group but significant in all patient groups (SD, $p<0.0001$, $t=7.45$; bvFTD $p<0.01$, $t=3.15$; AD $p<0.01$ $t=3.94$; PNFA, $p=0.04$, $t=2.66$). Additionally, subtracting the mean response to meaningless from that to meaningful sounds to produce a mean difference measure, the magnitude of this difference was significantly different between groups (initial one-way ANOVA $p<0.01$, $F=3.74$, $r2=0.21$) with greater responses in the SD group than healthy individuals ($p<0.01$, $F=3.74$, $t=2.91$, $r2= 0.21$). In those groups showing a deficit on the auditory semantic matching task (bvFTD, SD, AD), the magnitude of this difference also correlated with auditory semantic impairment (bvFTD $p<0.01$, $F=8.28$, $r2=0.78$, $t=-4.58$,; SD $p=0.02$, $F=7.91$, $r2=0.77$, $t= -3.02$; AD, $p=0.01$, $F=42.8$, $r2=0.97$, $t=-4.44$) but not with general disease severity markers (figure 6.2) (as indexed by symptom duration and reverse visual spatial span).

VBM co-ordinates are shown in table 6.3. Overall pupil reactivity to sound was significantly positively correlated with grey matter in the region of superior colliculus, whereas

the magnitude of the difference in pupil responses to M+ versus M- sounds was significantly inversely correlated with grey matter in left anterior superior temporal cortex (Figure 6.3; thresholded at $p<0.05$ after correction for multiple comparisons within regional anatomical volumes of interest).

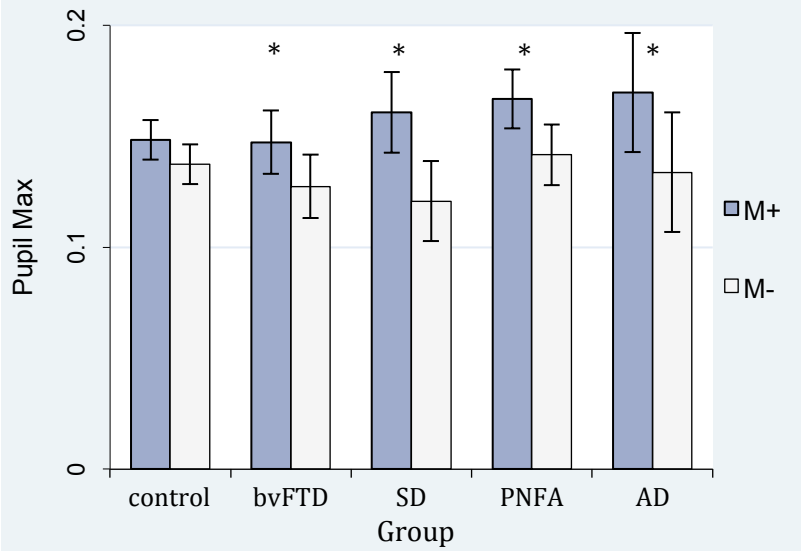

Figure 6.1. Mean maximal pupil dilatation response to sounds (Pupilmax): **A**, comparing real (meaningful, M+) and synthetic (meaningless, M-) sound conditions in each experimental group (*indicates significant difference between conditions, $p<0.05$; standard error bars shown).

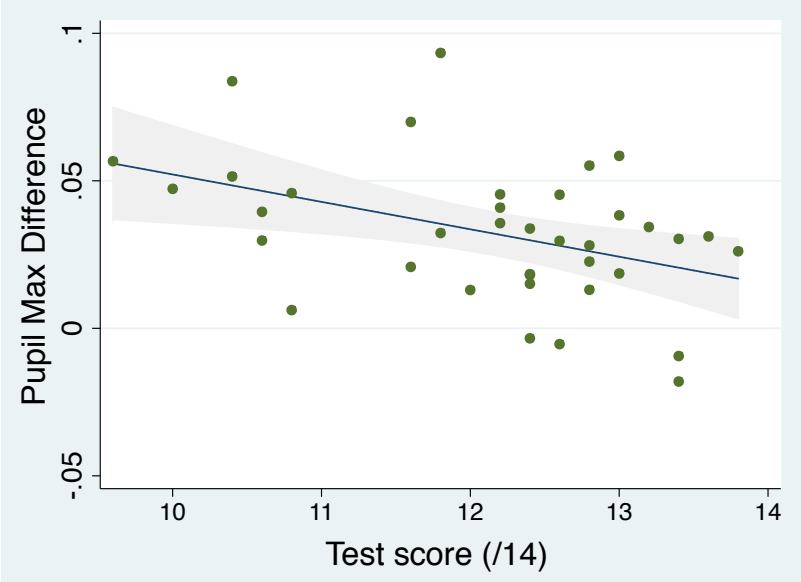

Figure 6.2. The magnitude of the difference in Pupilmax by sound condition (M+ minus M-) for each individual against their auditory semantic matching test score for the sounds used in the experiment. (linear regression best fit with 95% confidence intervals shown in grey).

Phenomenon	Group grey matter correlate	Brain region	Side	Local max (mm)			Z-score	Cluster size
				x	y	z		
overall pupil reactivity	diminished pupil reactivity associated with loss of grey matter	optic tectum	bilat	-12	-27	-6	4.12	499
response to M+ minus M- sounds	greater difference associated with grey matter loss	temporal pole	L	-51	9	-8	4.29	74

Table 6.3. Grey matter regions associated with key experimental parameters in the voxel-based morphometry analysis of the combined patient cohort are shown, together with coordinates of local maxima in MNI standard stereotactic space with associated Z-scores, and cluster sizes (number of voxels). Maxima shown were significant at threshold $p<0.05$ corrected for multiple comparisons within anatomical small volume of interest, based on prior hypotheses (see text). M+ meaningful (real) sounds; M- meaningless (synthetic) sounds.

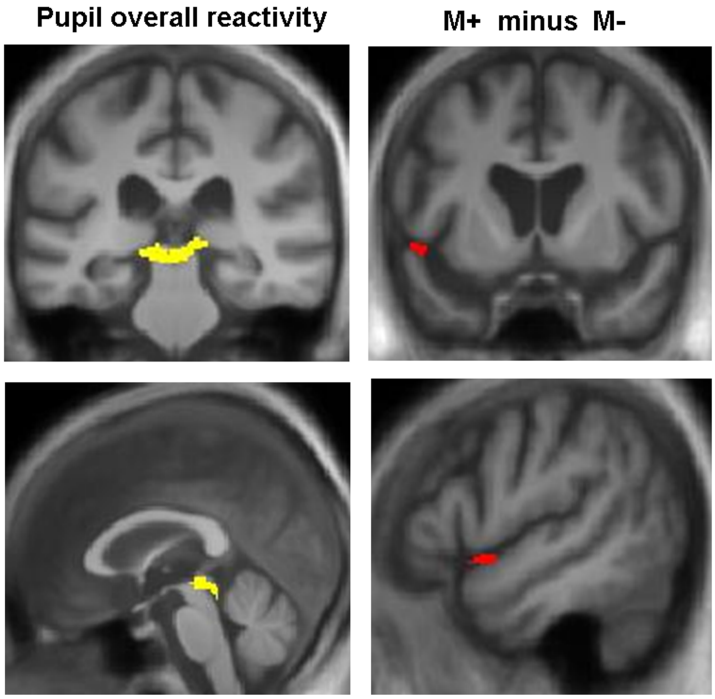

Figure 6.3. Overall pupillary reactivity (left) and difference in pupillary reaction to meaningful (M+) and meaningless (M-) sounds correlated with regional grey matter atrophy using statistical parametric mapping (all voxel-wise associations significant at $p<0.05$ after multiple comparisons correction within anatomical regions of interest shown rendered on coronal (above) and sagittal (below) sections. A positive grey matter correlate of overall pupil reactivity to sound was identified in superior colliculus (yellow); and an inverse correlate of the magnitude of the difference in mean Pupilmax to M+ over M- sounds in left anterior superior temporal cortex (red).

6.5 Discussion:

These findings show that autonomic responses as measured by pupil dilatations are modulated by the semantic content of nonverbal sounds in patients with damaged semantic systems. This effect on pupil responses was proportional to degree of auditory semantic impairment and most evident in patients with semantic dementia, the paradigmatic disorder of the human semantic system; the effect was not attributable to general autonomic or disease severity effects.

A priori it was hypothesized that the semantic content of a stimulus would act as a potent salience driver of autonomic responses and would evoke greater physiological reactions in healthy older adults than perceptually matched but 'meaningless' sounds. Here I observed only a trend towards greater responses in the latter.

In the SD group the responses were amplified towards meaningful sounds and appeared relatively depressed towards meaningless sounds, and although there were no significant difference in pupillary responses relative to controls when comparing either sound condition separately, the magnitude of these differences was significantly greater than in the control group.

So how do we interpret the exaggeration of pupillary reactions observed here in response to meaningful, and the depression of pupil responses in response to meaningless sounds in correlation with an impaired sematic system? The most likely explanation is that in addition to the salience effect of meaningfulness, sounds with a degree of semantic ambiguity are inherently salient. The biological purpose for this is clear; a sound where the source is not immediately apparent, especially in situations where additional contextual information to aid identification (for example situations where other sensory input is degraded such as in darkness with 'things that go bump in the night'), require more attention and cognitive processing for source identification. Sounds with high semantic content that are inherently less ambiguous, require less cognitive processing effort to identify and pupillary response here is not driven so much by

cognitive workload related to ambiguity but by biological relevance; hence why in this experiment 'real' sounds evoke larger pupillary responses than the less biologically relevant synthetic sounds. This would, in a simplified manner, result in a Gaussian relationship between semantic content of the stimuli and pupillary response as shown in figure 6.4. If the semantic system is degraded then this normal relationship between pupil response and semantic content could potentially shift to the left. Sounds of high semantic content (A) will be rendered more ambiguous requiring greater cognitive processing and evoking greater pupillary responses. Sounds that normally lie to the left of the middle (B), i.e. are of lower semantic content (the meaningless sounds here), in the context of a deranged semantic processing system will be rendered effectively 'irrelevant, of little cognitive interest, and will evoke little pupillary response.

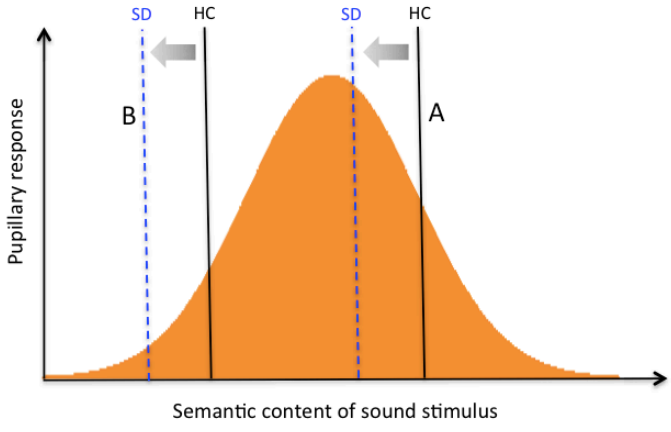

Figure 6.4. cartoon of a possible mechanism to account for the greater pupillary responses observed for high semantic content sounds (A) and lower pupillary responses to low semantic content sounds (B) in SD relative to healthy controls.

A gradation of the relationship between the semantic component of the stimulus and pupillary response could be more clearly mapped with the use of stimuli graded for semantic content (rather than binarised). Alternatively, manipulating the context in which sounds are presented would alter the degree of ambiguity by shifting the most likely explanation for a given stimulus. Further work could investigate the relationship of semantic and salience using

vocalisations; a sound category likely on evolutionary grounds to be highly salient. Language would be an obvious way of manipulating semantic content of speech whilst maintaining other factors such as emotional content, whereas manipulation of animal vocalisations to perceptually matched but meaningless counterparts may lend themselves to the investigation of relative effects of semantic versus perceptual cues on semantic processing. As we saw in chapter 4 'primitive' cues such as motion are strong salience drivers and it may well be that sounds with the spectral shape of animal calls but artificial and unidentifiable may evoke similar responses.

The correlation of right anterior temporal pole atrophy with the magnitude of difference in response to M+ minus M- sounds is consistent with previous neuroimaging evidence implicating the non-dominant anterior temporal lobe in nonverbal sensory semantic processing of sounds and other sensory objects (Goll *et al.*, 2010; Lambon Ralph *et al.*, 2010; Omar *et al.*, 2010; Hsieh *et al.*, 2011; Visser & Lambon Ralph, 2011; Omar *et al.*, 2013). Further, the right anterior temporal lobe is considered a key 'hub' in an 'appraisal' network that links closely with the salience network (Seeley *et al.*, 2007b; Seeley *et al.*, 2009; Guo *et al.*, 2013); inappropriate (semantically driven) appraisal of auditory stimuli may result in over attribution of salience.

Although the VBM findings must be interpreted with a degree of caution as the methodology used risks the production of type-1 error, as discussed in chapter 3, the finding of the involvement of the superior colliculus in relation to overall pupillary reactivity is of interest. The SC has been long known to be involved in orienting response (Kustov & Robinson, 1996; Lunenburger *et al.*, 2001; Wang *et al.*, 2012; Krauzlis *et al.*, 2013; Mysore & Knudsen, 2013; Wang *et al.*, 2014) and via the thalamus mediates motor output to the eye, head and neck, and arm and shoulder from cortical areas including the frontal eye fields. Stimulation of the SC results in coordinated head and eye gaze shifts (Freedman *et al.*, 1996) (Freedman et al., 1996) and more recently, it has been shown that stimulation at thresholds below those necessary to evoke saccadic eye movements results in pupillary dilatation in both monkeys and owls (Netser *et al.*, 2010; Wang *et al.*, 2012). It may therefore be that the correlation of superior colliculus atrophy

and decreased pupillary responses observed here reflects impaired effector pupil control mechanisms. However, single cell recordings from the superior colliculus show correlations with the salience of the stimuli presented with the superior colliculus suggested to play a key role in salience processing (Wang *et al.*, 2012) and it may therefore be that impaired pupillary responses observed reflect impaired core salience processing.

6.6 Chapter conclusions:

Impaired non-verbal auditory semantic ability leads to an exaggeration of a normal trend towards greater physiological reactions to meaningful relative to matched meaningless sounds. The magnitude of these differences correlated with the degree of auditory semantic impairment and may reflect the increased cognitive processing demands of ambiguity analysis by an impaired semantic system.

The magnitude of these differences differentiate patients with SD from the healthy older controls, which may provide avenues for further physiological biomarker development. The degree of semantic impairment had a right anterior temporal lobe correlate suggesting that much salience information is semantically mediated and underpinned by right anterior temporal lobe function.

Chapter 7: Abnormal hedonic processing of sound and music in FTD and AD

7.1 Chapter Summary:

Symptoms found in FTD are suggestive of abnormal hedonic processing. Here I investigate these in relation to abnormal sound and music hedonic coding and correlate to underlying neuroanatomical substrates. Symptoms were found to be common reflecting changes in both sound and music appreciation and were present in FTLD and AD, although the prevalence and directionality of the changes differed between groups. Further, symptoms suggestive of abnormal hedonic processing were shown to dissociate by modality with increased pleasure for music occurring in associating with decreased pleasure from environmental sounds. Neuroanatomically, symptoms were associated with atrophy of areas implicated in reinforcement learning and emotion processing circuitry as well at the right anterior temporal lobe, suggesting a role for deranged contextual evaluation. Together, these findings provide further evidence that symptoms observed in dementia are underpinned by abnormalities to key physiological processing systems.

7.2 Introduction:

What makes something pleasant and what happens when these processes go wrong? Examination of sound and music hedonic processing may help us shed light upon these questions. In FTD insidious and progressive alterations of behaviour are common, especially in bvFTD and SD (Edwards-Lee *et al.*, 1997; Snowden *et al.*, 2001; Hodges & Patterson, 2007; Rankin *et al.*, 2009; Rohrer & Warren, 2010b; Kumfor *et al.*,

2011; Rohrer & Warren, 2011; Duval *et al.*, 2012; Rohrer *et al.*, 2012; Clark *et al.*, 2014a) and in addition to primary deficits in emotion processing pathway, it has been suggested that some of these behaviours, in particular binge eating, hypersexuality, and new drug and alcohol use may reflect derangement of normal reinforcement learning processing pathways (Miller *et al.*, 1995; Cruz *et al.*, 2008; Mendez & Shapira, 2013; Perry *et al.*, 2014; Perry & Kramer, 2015). Key cortical and subcortical brain regions implicated in the processing of positive reinforcement overlap those bearing the brunt of disease in FTD (Whitwell *et al.*, 2007; Woolley *et al.*, 2007; Seeley *et al.*, 2009; Piguet, 2011; Halabi *et al.*, 2013; Sescousse *et al.*, 2013; Moller *et al.*, 2014); aberrant reinforcement learning processing in FTD is therefore anticipated from neuroanatomical grounds.

As well as symptoms in FTD suggestive of deficient reinforcement learning processing of more ecological positive reinforcer types, symptoms emerge that speak to aberrant hedonic processing of another key stimulus, an abstract stimulus that for many carries great pleasure, despite no obvious biological purpose; namely music. Music produces strong physiological and psychological arousal and listening to music can be one of the most pleasurable experiences known to humans (Krumhansl, 1997; Sloboda, 2001; Khalfa *et al.*, 2002; Baltes *et al.*, 2011). The fact that music is ubiquitous across societies and has evolved from pre-Neolithic times alongside the rest of human civilization is testimony to its importance to mankind (McDermott, 2008). Despite this, however, unlike many positive reinforcers such as food and sex, it is an abstract concept serving no obvious inherent purpose (McDermott & Hauser, 2004; McDermott & Hauser, 2007). Studies of hedonic and positive reinforcer processing of music suggest that the underlying brain circuitry is common to that implicated in positive reinforcement of more biologically germane sensory stimuli (Blood *et al.*, 1999; Blood & Zatorre, 2001; Menon & Levitin, 2005). However, 'musical anhedonics', healthy individuals who do not gain pleasure or mount physiological responses to music, despite normal responses to monetary positive reinforcer have also recently been described

(Mas-Herrero *et al.*, 2014), suggesting the existence of music-specific positive reinforcement systems (Clark *et al.*, 2014c). If the neurobiological systems underpinning positive reinforcement of music versus more generic positive reinforcement stimuli can become uncoupled, this may offer avenues for exploration of brain mechanisms of positive reinforcement learning in health and disturbances of these in disease.

Musical pleasure likely depends upon factors such as recognition, familiarity, and coherence to pre-learned musical 'rules' on pattern evolution predictions based upon individual musical experience (Zatorre & Salimpoor, 2013; Koelsch, 2014; Salimpoor *et al.*, 2014); anatomically based upon temporo-frontal networks that are particularly affected in FTD and derrangement of music hedonic processing seems likely. However this remains relatively under-explored. I recently demonstrated the development of abnormal musical hedonic evaluation ("musicophilia") in FTD in association with relatively preserved left hippocampal grey matter volume relative to those without the phenomenon (Fletcher *et al.*, 2013). However, in this study sample sizes were small and whether the hedonic value of music could be decreased, producing a musical anhedonia, or even rendering music aversive, has not been explored. Further, impairment of perceptual encoding of environmental sounds has been demonstrated in FTD, with both derangement of basic feature representation, or disruption of semantic encoding demonstrated (Goll *et al.*, 2010). Whether derangement of the hedonic evaluation of environmental sounds also occurs has not been established and on neuroanatomical grounds, any changes observed in musical positive reinforcer processing may reflect disruption of a more generalised auditory-positive reinforcer processing system. If the extraction of positive reinforcer value from ecological stimuli such as environmental sounds, and the more abstract stimulus of music, are underpinned by overlapping but dissociable neuroanatomical substrates, then different disease processes may carry different behavioural and anatomical signatures and may dissociate from abnormalities of primary positive reinforcer processing; characterising

these behaviours and the neural underpinning is clearly of neurobiological interest and clinical relevance.

Hypotheses and predictions: Here I examined in a large cohort of patients with FTD in comparison to AD the characteristics and underlying neuroanatomical correlations of the evolution of musicophilia, musical aversion and a generalised environmental sound aversion in relation to changes in primary positive reinforcer processing, namely the evolution of a sweet-tooth. I hypothesised that changes in sound pleasure behaviours are relatively common, can dissociate by sound type and from other types of reinforcer, and are underpinned by damage to brain regions involved in normal reinforcement learning such that emotional salience processing is disrupted.

7.3 Methods:

7.3.1 participant characteristics:

Seventy-three patients with dementia participated in this study; 56 patients with a syndrome of FTLD (bvFTD, n=22; SD, n=19; PNFA, n=15) and 17 patients with amnestic AD. CSF or brain amyloid PET imaging findings were in keeping with syndromic diagnosis (ratio of total tau: beta-amyloid$_{1-42}$ levels >1 in 9/9 AD patients and <0.8 in 14/14 FTLD patients, Flubetapir PET negative for amyloid deposition in 6/6 FTLD patients for whom data were available). Genetic screening of the patient cohort revealed 13 patients with a pathogenic mutation (seven C9orf72; six MAPT). All patients with a genetic mutation presented with bvFTD apart from one patient with a C9orf72 expansion who presented with PNFA. In order to minimize potential confounds, participants with peripheral hearing loss (n=12) or tinnitus (n=8) were excluded.

7.3.2 Analysis of hedonic symptoms:

Patient caregivers were asked to complete a questionnaire detailing any symptoms suggesting alterations in the pleasure the subjects derived from environmental sounds and/or music (see Table 7.1). Alterations in hedonic responses were classified broadly as increased or decreased liking for environmental sounds and increased or decreased liking for music. Altered liking was referenced to the patient's premorbid behaviour as reflected in expressed liking or aversion for the sound, seeking or avoidance of the sound and/or amount of time spent listening to music. The caregiver questionnaire also recorded any alteration in patients' sweet food preference, in order to assess altered hedonic valuation of sounds in relation to another hedonic behaviour that is commonly affected in dementia (Woolley *et al.*, 2007; Perry *et al.*, 2014).

Patient subgroups with and without hedonic symptoms and healthy controls were compared using linear regression and proportions exhibiting symptoms, and the presence of any correlation between auditory and food preference alterations in each patient subgroup were compared using Pearson's chi-square. A threshold $p<0.05$ was accepted as the criterion for a statistically significant difference in all comparisons.

Has he/she become more sensitive to sound than before the illness? If yes, please give details:
Does he/she seem to find some sounds more pleasant or less pleasant than before the illness? If yes, please give details:
Has his/her appreciation of music altered compared with before the illness? If yes, please give details:
Has his/her liking for sweet foods altered compared with before the illness? If yes, please give details:

Table 7.1. questions administered to carers probing changes in music, sound and food preference

7.3.3 Brain image acquisition and voxel-based morophometry:

At the time of questionnaire data collection each patient underwent volumetric brain MRI using the methodology descried in chapter 2. Using linear regression, voxel intensity (grey matter volume) was modelled over the FTD and the AD cohorts separately as a function of presence of symptoms suggestive of any alteration in sound pleasure and additionally, as a function of altered liking for music or environmental sounds in isolation and altered liking for sweet foods. Anatomical small volumes of interest based on the prior anatomical hypotheses were created to cover key areas previously implicated in hedonic processing of sounds and other sensory stimuli (medial temporal lobe structures covering hippocampus and amygdala, anterior cingulate, insula cortex and striatum: (Sescousse *et al.*, 2013)). As likely lateralisation of any observed effects could not be predicted a priori, regions of interest were examined for both hemispheres separately. This yielded a total of 7 small volume corrections (the striatal small volume covered both hemispheres).

	Healthy controls*	FTLD		AD		ANOVA
		no auditory hedonic a	auditory hedonic b	no auditory hedonic c	auditory hedonic d	
General						
No. (F:M)	50 (23:27)	25 (15:10)	31 (9:22)†	10 (5:5)	7 (2:5)††	
Syndrome: bvFTD/SD/PNFA	NA	03/08/2014	19/11/2001	NA	NA	
Genetic: C9orf72 / MAPT	NA	4/0	03-Jun	NA	NA	
Age (years)	67.5 (54-80)	64.9 (52-75)	64.7 (52-79)	66.4 (53-80)	66.3 (60-73)	
Education (years)	15.2 (10-18)	15 (11-20)	14 (11-21)	13.9 (12-17)	13 (11-17)	
Symptom duration (years)	NA	5.7 (3-21)	6.1 (3-18)	4.4 (2-6)	6 (4-9)	
MMSE	29.6 (28-30)	**19 (1-30)**	**24 (12-30)**	**20 (13-30)**	24 (20-25)	p=0.001, F=6.6, r^2= 0.27
IQ						
Verbal	120 (101-137	**82 (55-115)**	**80 (40-119)**	**84 (55-108)**	**94 (71-115)**	p<0.0001, F=36.39, r^2=0.6
Performance	115 (84-141)	**96 (66-135)**	**101 (74-135)**	**84 (57-119)[a,b]**	**86 (61-125)**	p<0.0001, F=13.70, r^2=0.35
Episodic memory						
RMT faces (/50)	43 (30-50)	**37 (25-46)**	**31 (24-50)[a]**	**32 (23-46)**	**36 (27-43)**	p<0.0001, F=20.30, r^2=0.44
RMT words (/50)	48 (39-50)	**37 (18-48)**	**34 (23-49)**	**31 (27-42)[a]**	**34 (27-47)**	p<0.0001, F=45.84, r^2=0.63
Semantic processing						
BPVS (/150)	147 (137-150)	**109 (25-149)**	**110 (2-149)**	**124 (52-147)**	133 (106-146)	p<0.0001, F=9.28, r^2=0.24
Executive function						
Stroop inhibition (180 sec)	58 (35-103)	**103 (50-180)**	**88 (40-180)**	**107 (73-138)**	**135 (42-180)[b]**	p<0.0001, F=12.91, r^2=0.37
Digit span reverse (/12)	5 (3-7)	**4 (0-7)**	**4 (0-7)**	**3 (1-5)[b]**	**3.5 (2-6)**	p<0.0001, F=9.02, r^2=0.26
Visuospatial						
VOSP object decision (/20)	18 (12-20)	**16 (3-20)**	**16 (8-20)**	**16 (11-19)**	16.5 (14-18)	p<0.01, F=3.99, r^2=0.13

Table 7.2. General demographic and neuropsychological data for patient subgroups with and without auditory hedonic symptoms. Mean (range) data are shown unless otherwise indicated and maximum scores on neuropsychology tests are also indicated in parentheses. Initial one-way ANOVA prior to specific group comparisons is displayed in the right hand column and significant differences (p<0.05) between patients and healthy controls are in bold; *historical control group (to reference neuropsychological characterisation of disease groups); †four patients with environmental sound aversion alone, 10 with musicophilia alone, eight with music aversion alone, five with both musicophilia and environmental sound aversion, four with both music aversion and environmental sound aversion (see text, Figure 7.1); ††five patients with environmental sound aversion alone, two with both music aversion and environmental sound aversion; [a]significantly (p<0.05) different from non-symptomatic patients with FTLD; [b]significantly (p<0.05) different from non-symptomatic patients with AD

7.4 Results:

7.4.1 General participant characteristics:

Demographic, clinical and general neuropsychological characteristics of the patient cohort are summarised in Table 7.2. Participant subgroups (FTLD versus AD and within each disease group, subgroups with and without altered sound pleasure) did not differ in age, gender, years of education, disease duration or overall severity (based on MMSE score). On general neuropsychological assessment the FTLD subgroup with auditory hedonic symptoms performed significantly worse on the recognition memory test for faces than the FTLD subgroup without such symptoms; there were no other significant neuropsychological differences between disease subgroups with and without auditory hedonic symptoms.

7.4.2 Characteristics of hedonic symptoms:

Symptoms of altered auditory hedonic valuation occurred in a substantial proportion of patients in both the FTLD cohort (31/56 cases, 55%) and the AD cohort (7/17 cases, 41%), with no statistical difference in frequency in each disease. The breakdown of auditory hedonic symptoms by diseases and syndromes is schematised in Figure 7.1. Within the FTLD cohort, symptoms were significantly more common in the bvFTD group (19/22 cases, 86%) than the SD group (11/19 cases, 58%) ($p=0.04$, $\chi^2=4.21$) and in both the bvFTD and SD groups than the PNFA group (1/15 cases, 7%) (bvFTD vs. PNFA, $p<0.0001$, $\chi^2=22.81$; SD vs. PNFA $p<0.01$, $\chi^2=9.63$). Altered liking for environmental sounds and for music were each exhibited by patients in both the FTLD and AD cohorts, however the relative frequency and directionality of these symptoms varied between diseases: patients with FTLD who developed auditory hedonic symptoms variously exhibited decreased liking for environmental sounds (13/31 cases, 42%), decreased liking for music (12/31 cases, 39%) or increased liking for music

('musicophilia'; 15/31 cases, 48%) alone or in combination, whereas those patients with AD who developed hedonic symptoms uniformly exhibited decreased liking for sounds (environmental sounds in 7/7 cases; music additionally in 2/7 cases, 29%). Only one patient in the entire study cohort experienced abnormally increased liking for neutral environmental sounds: this patient with a syndromic diagnosis of SD derived pleasure from certain mechanical sounds such as a hair-dryer as well as exhibiting a heightened emotional response to music.

Caregiver questionnaire reports indicated a range of descriptions of altered auditory hedonic responses in individual patients (representative extracts for individual patients are in Table 7.3). Typically patients with reduced liking for environmental sounds were described by caregivers as having become unusually sensitive to the relevant sound since onset of their illness; for example, certain environmental noises (particularly those with higher pitch or penetrating timbre such as children's voices) would provoke expressions of distress and they would take sometimes elaborate steps to avoid such sounds, even in situations where these would previously have been regarded as unobtrusive or banal. Patients with reduced liking for music were described as exhibiting a wider range of responses, from indifference (loss of previous interest and enjoyment) to active avoidance, distress or irritation; I therefore refer to environmental sound and music 'aversion' to capture the range of responses. Conversely, patients with increased liking for music (in line with previous descriptions of musicophilia: (Fletcher *et al.*, 2013)) exhibited music craving or seeking, often demanding to listen to a narrow repertoire of songs for up to many hours each day but sometimes also engaging in more organised behaviours such as taking up a musical instrument or buying music equipment.

Within the FTLD subgroup with altered auditory hedonic responses, patients with bvFTD and SD were comparably likely to develop environmental sound aversion (bvFTD 9/19 cases, 47%; SD 4/11 cases, 36%); there was the impression of an over-

representation in the SD group of patients with musicophilia (bvFTD 8/19 cases, 42%; SD 7/11 cases, 64%) versus music aversion (bvFTD 8/19 cases, 42%; SD 3/11 cases, 27%), however this apparent disproportion did not achieve statistical significance when the SD and bvFTD groups were compared directly. Interestingly, the directionality of the alterations in hedonic responses could dissociate with musicophilia evolving in conjunction with environmental sound aversion in a substantial minority of patients with FTLD (bvFTD 3/19 cases, 16%; SD 2/11 cases, 18%); a comparable proportion of patients (6/38 cases, 16% of the combined cohort) exhibited aversion to both sound categories. The single patient with PNFA who developed auditory hedonic symptoms exhibited music aversion. Genetic FTLD subtype influenced the development of auditory hedonic alterations: symptoms were significantly more common in the MAPT mutation group (6/6 cases) than the C9orf72 mutation group (3/7 cases) ($p=0.03, \chi^2=4.95$). Patients in both these genetic subgroups tended to exhibit aversion to sounds; musicophilia was reported only in isolated cases in each subgroup (in each case accompanied by environmental sound aversion).

Compared with auditory hedonic symptoms, pathological sweet–tooth developed in a similar proportion of the FTLD cohort overall (35/56 cases, 63%) and in association with bvFTD (21/22 cases, 95%) and SD (9/19 cases, 47%). Development of pathological sweet tooth was significantly correlated with development of any auditory hedonic symptoms in both the FTLD and AD groups (FTLD, $p<0.0001, \chi^2=23.16$; AD $p<0.001, \chi^2=14.78$) but not more specifically with a particular auditory hedonic phenotype. Again, like within the auditory domain, increased liking for sweet foods dissociated from changes in sound hedonic responses with both increased and decreased sound pleasure responses reported. Development of auditory hedonic symptoms or pathological sweet tooth were not significantly correlated with disease duration or severity (MMSE score), in either FTLD or AD.

7.4.3 Neuroanatomical associations:

Regional grey matter correlates of auditory hedonic symptoms from the VBM analysis are summarised in Table 7.4 and statistical parametric maps are shown in Figure 7.2.

At the most stringent statistical criterion ($p<0.05_{FWE}$ corrected over the whole brain volume), within the combined FTLD cohort the presence of any auditory hedonic symptoms was associated with grey matter loss in right temporal pole and anterior superior temporal cortex, extending into mid and posterior insula and putamen; while the presence of environmental sound aversion alone was also associated with grey matter loss in right anterior temporal lobe and insula, extending to include right amygdala, hippocampus, entorhinal and parahippocampal cortex.

No other grey matter associations of auditory hedonic symptoms were identified at whole brain level. However, further neuroanatomical associations were identified in the FTLD cohort at significance threshold $p<0.05_{FWE}$ corrected within the anatomical regions specified by my prior hypotheses. At this criterion, environmental sound aversion was associated with additional grey matter loss in left amygdala and nucleus accumbens. Music aversion was associated with grey matter loss in an overlapping network including right anterior temporal cortex, entorhinal cortex, hippocampus and amygdala and bilateral mid and posterior insula. No neuroanatomical associations of musicophilia were identified at the prescribed significance threshold; however, a post hoc analysis at a more lenient threshold ($p<0.001$ uncorrected over the whole brain) revealed relative preservation of grey matter in right hippocampus (MNI peak coordinates [38 -1 -28], z-score 3.80) in association with musicophilia.

In the AD cohort, the presence of environmental sound aversion was associated with grey matter loss in anterior cingulate cortex at significance threshold $p<0.05_{FWE}$ corrected within the pre-specified anatomical region of interest.

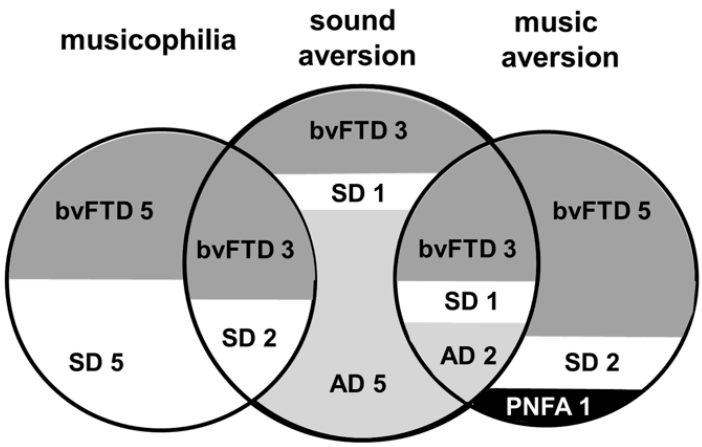

Figure 7.1. Breakdown of auditory hedonic symptoms across the patient cohort. Case numbers in each symptom category are indicated.

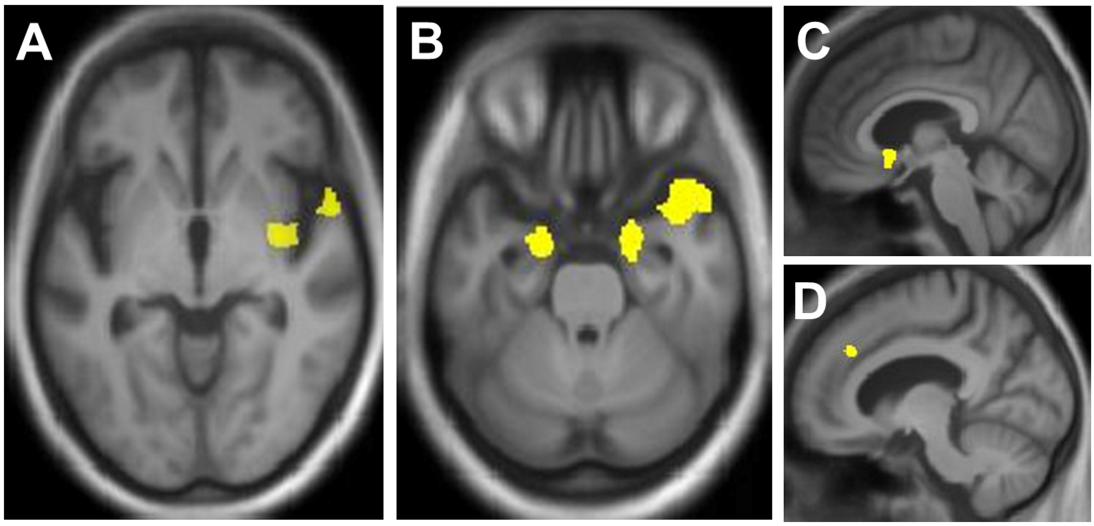

Figure 7.2. SPMs showing regional grey matter atrophy significantly associated with: **A**, any auditory hedonic symptoms in the combined FTLD cohort, centred on right anterior temporal lobe, insula and putamen; **B**, **C**, environmental sound aversion in the combined FTLD cohort, including anterior temporal cortex, amygdala and nucleus accumbens; **D**, environmental sound aversion in the AD cohort, in anterior cingulate cortex. The neuroanatomical associations of music aversion in the FTLD cohort (not shown) comprised a similar distributed fronto-temporal network (see Table 7.4). SPMs are thresholded at $p<0.05$ after small volume correction for multiple voxel-wise comparisons in pre-specified small anatomical volumes of interest

Case	Sub-group	Comment
1	bvFTD: C9orf72	Seems sensitive to loud music. Only listens to music for short periods of time now.
2		More sensitive to any noise. Doesn't like listening to music any more.
3		Increasingly sensitive to noises, in particular children's voices he finds unpleasant and agitated by these. Listens to i-pod constantly now loves music, plays it very loudly even throughout the night
4	bvFTD: MAPT	Doesn't really like music any more as it gives no pleasure
5		Plays less music at home, doesn't enjoy it anymore
6		Used to sing in choir and play in orchestra, now never puts on music to listen to at home any more.
7		Does not like any loud noises of any sort, easily agitated by fire alarms, helicopters overhead, sirens
8		Doesn't like loud bangs or crashes which never used to bother him before.
9		Notices sounds from neighbours or planes more than others do, irritated by the sound of furniture scraping on the floor. Listens to music all the time now, same records over and over
10	bvFTD: sporadic	More sensitive to sound. Used to love classical music but now turns the radio off, even when I am listening, doesn't enjoy music any more
11		Will sit and watch music videos constantly on the music channel for hours at a time
12		Likes music more, has bought a juke box which he plays a lot and has started trying to re-learn the piano! Emotional in response to music
13		Now likes complete silence without radio or TV; children's voices are particularly unpleasant and will sit as far away as possible from the grandchildren when we visit
14		Can't stand certain sounds, especially the sound of birds tweeting or young children's voices, engine sounds. But really enjoys music, and has started trying to play the piano and sing (loudly) all the time
15		Hearing seems to have been heightened. Constantly buying music CDs, obsessively uploads music to i-pod, plays it very loudly
16		Loud sounds now generally upsetting but plays music very loudly 24 hours a day now.
17		Likes music more
18		Loud sounds are distressing and jumps very easily if there is a sudden loud noise. Background music causes huge irritation and distress
19		Hypersensitive to noise on the train will move away from people who are talking even if it means standing, won't have the gas fire on (it 'pops'). Demands music on radio turned off even in other room.
20	SD	No interest in listening to music now, less appreciative of any music.
21		Loves all kinds of music more than before
22		Likes listening to music more, dances along
23		Seems to enjoy loud sounds even if non-musical, the new hand driers give auditory pleasure. More aware of music and gets more emotional
24		Finds loud noises more irritating
25		Real dislike of noises now, even if not particularly loud, especially children's voices, trains, sirens, reduced to tears by sound of a fast train passing through station Likes music more; now obsessed with music videos from 40s and 50s, watches these >50 times a day, has also begun picking out the same tunes on the piano (last played 25 years ago). Obsessional, missed Christmas lunch to do this.
26		Always had the radio or a CD on, now hardly bothers listening to the radio and will never play music
27		Wants to play music more and louder, likes music more now.
28		Listens to music a lot more now
29		Leaves the room if coffee ground or food processor on. Doesn't enjoy listening to music like before
30		Very upset by loud noises, e.g. passing trains, hairdryers, children's voices. Likes music more - obsessed with particular 1950s singers
31	PNFA	Does not enjoy listening to music any more though previously enjoyed a wide range
32	AD	More irritated by noises like TV and radio than before
33		Complains TV and radio too noisy when they are not, finds voices more irritating
34		Does not like the sound of the telephone ringing now
35		Seems more sensitive to loud sounds, finds voices unpleasant
36		Now seems to find Big Ben tolling on the news and similar tones quite excruciating, finds high pitched sounds in films unbearable. Used to enjoy rock music but now finds most music just irritating noise
37		Less tolerant of everyday sounds, finds them more irritating (e.g., some recent not particularly loud building work nearby caused distress)
38		Hearing seems to have become more sensitive especially to high pitched noises like children screaming. Now finds music irritating

Table 7.3. Representative care giver comments for patients with auditory hedonic symptoms

Auditory hedonic symptom	Brain region	Side	Cluster size (voxels)	Co-ordinates (mm)			z-score
				x	y	z	
FTLD							
Any hedonic alteration	Anterior superior temporal gyrus	R	3002	54	15	-9	**4.86**
	Anterior superior temporal sulcus	R		51	5	-15	4.43
Environmental sound aversion	Anterior temporal cortex	R	5275	42	17	-26	**4.88**
	Nucleus accumbens	R	668	3	15	-9	4.24
	Amygdala	L	413	-20	-3	-26	3.66
	Inferior temporal gyrus	R	209	50	0	-39	3.72
Music aversion	Hippocampus/ amygdala	R	2864	26	-25	-24	4.69
	Mid – posterior insula	R		44	-1	-12	4.20
	Entorhinal / parahippocampal cortex	R		24	2	-35	4.10
	Mid – posterior insula	L	128	-42	-9	-5	3.83
AD							
Environmental sound aversion	Anterior cingulate cortex	L	87	-12	32	31	4.21

Table 7.4. Neuroanatomical associations of hedonic symptoms in the patient cohort. Regional grey matter associations shown were all significant at threshold $p<0.05_{FWE}$ corrected for multiple comparisons within the pre-specified anatomical small region of interest; associations in bold were additionally significant at $p<0.05_{FWE}$ corrected over the whole brain volume. All associations are with grey matter atrophy except where designated (relative grey matter preservation associated with musicophilia)

7.5 Discussion:

Here I demonstrate that alterations in music and environmental sound hedonic processing occur commonly in both FTD and AD, with the pattern varying by syndrome; whilst patients with FTD showed bidirectional change in music appreciation, patients with AD developed an environmental sound aversion, including an additional aversion to music in two cases. Within the FTD group, dissociation between hedonic music and environmental sound processing was also demonstrated, with the presence of increased musical pleasure and obsession in association with distress or irritation with usually innocuous environmental sounds. Abnormal hedonic processing was over-represented in the MAPT group (6/6 subjects). Changes in positive reinforcer processing for environmental sounds and music also dissociated behaviourally from processing of the primary positive reinforcer of food.

Within the FTD cohort, the presence of any alteration in sound hedonic valence was associated with grey matter loss in the right anterior temporal lobe extending into the middle and posterior insula and putamen. The presence of an environmental sound aversion was also associated with atrophy of the right anterior temporal lobe and insula but in this case extending throughout the mesial temporal lobe structures to include the right hippocampus, and amygdala. After examination of a priori defined regions of interest, atrophy was also found in association in the left amygdala and nucleus accumbens. An overlapping network was observed in association with the evolution of a musical anhedonia/music aversion: here atrophy was observed in the right mesiotemporal structures; throughout the hippocampus extending into the amygdala, as well as regions bilaterally in the insula cortex. Musicophilia was associated with relative preservation of the right hippocampus. Within the AD group the presence of symptoms was associated with atrophy of the left anterior cingulate cortex.

The findings are consistent with and extend previous work on reward processing in bvFTD demonstrating atrophy in the right putamen and fronto-insula cortex in association with

symptom of over-eating, sweet craving, hypersexuality and drug use (Whitwell *et al.*, 2007; Woolley *et al.*, 2007; Goll *et al.*, 2010; Perry *et al.*, 2014; Perry & Kramer, 2015).

Whilst the encoding of both basic perceptual sound characteristics and semantic processing of music and environmental sounds has been demonstrated to be impaired in FTD (Hailstone *et al.*, 2009; Goll *et al.*, 2010), I demonstrate here that changes in hedonic processing of sounds reflects dysfunction of areas well established in generalized reinforcement processing circuitry; areas that include the amygdala, hippocampus, nucleus accumbens, and insula cortex (Breiter *et al.*, 1997; Breiter & Rosen, 1999; Knutson *et al.*, 2001; Mobbs *et al.*, 2003; Robinson & Berridge, 2003; O'Doherty, 2004; Pessiglione *et al.*, 2006; Sescousse *et al.*, 2013).

The hedonic and positive reinforcement value of an environmental sound is likely to be heavily influenced by perceptual cues and semantic associations, and, as discussed in earlier chapters, these processes are disrupted in FTD, leading to abnormal cognitive evaluation and physiological responses towards emotionally laden sound. The cognitive evaluation of the positive reinforcement content of music is likely more complex, with less direct semantic associations and biological purpose. The positive reinforcer of music evolves over time as a piece unfolds, and is likely dependent upon coherence to temporal predictions that will be based upon individual musical experience (Zatorre & Salimpoor, 2013). Additionally, the positive reinforcement value of music increases with familiarity (Salimpoor *et al.*, 2013) and no doubt the elements that make any given piece of music appear pleasurable to our ear are multifactorial, influenced by brain systems involved in processes including those determining the evaluative emotional state of the listener, familiarity and explicit recognition of the musical piece (semantic memory underpinned by anterior temporal lobe networks), contextual associations (episodic memory of the mesial temporal lobe structures) and more executive functions including working memory, temporal sequencing, planning and expectation (pre-frontal regions), in addition to those systems involved in generalized positive reinforcer processing (Zatorre & Salimpoor, 2013; Koelsch, 2014).

This is highlighted by the robust finding at whole brain correction of involvement of the right anterior temporal lobe here, a region not generally considered part of the core reinforcement learning or emotional processing circuitry that was not included in the a priori regions of anatomical interest. In addition to key reinforcement learning areas (amygdala, ACC, striatum), the R ATL has strong interconnections with regions involved in interoceptive representation (posterior insula), and the emotionally mediated memory areas (hippocampus and parahippocampal formations) forming an 'appraisal network' (Guo *et al.*, 2013). This network is disrupted in SD and heavily overlaps a salience processing network, which is particularly vulnerable in bvFTD (Seeley *et al.*, 2007b; Seeley *et al.*, 2009). In addition, the right anterior temporal lobe acts as a key hub in processing of non-verbal multi-modal semantic information (Hailstone *et al.*, 2009) and lesions result in impairment of recognition of music emotion in FTD (Hailstone *et al.*, 2009; Omar *et al.*, 2010; Hsieh *et al.*, 2011; Omar *et al.*, 2011a; Hsieh *et al.*, 2012b). Lesions of the right anterior temporal lobe also result in de-novo somatisation behaviour and abnormal sensory behaviours (Snowden *et al.*, 2001; Chan *et al.*, 2009), which may reflect abnormal attribution of salience to normally innocuous stimuli (see chapter 3). Putting this together, the right ATL may play a role here in appraising the salience of incoming sensory information in a semantically appropriate context relative to previous autobiographical knowledge (Rankin *et al.*, 2006; Irish *et al.*, 2014); degradation of this system may lead to inappropriate contextualisation and mis-assignment of hedonic value to stimuli, in this case rendering usually pleasant or innocuous sounds aversive.

That amygdala involvement was found in association with the loss of the normal hedonic value of music (music aversion/anhedonia) and the development of an aversion to what are usually innocuous sounds (environmental sound aversion), accompanied by NAcc atrophy in the latter, is of interest. The amygdala and NAcc are functionally heavily interconnected (Murray, 2007; Haber & Knutson, 2010), and the strength of these connections increase functionally with increasing positive reinforcement value of music (Blood & Zatorre,

2001). These regions are not only implicated in multi-modal positive reinforcement processing (including the positive reinforcement from viewing beautiful faces, obtaining desirable objects, and more abstract concepts such as music, reputation and social hierarchy (Aharon *et al.*, 2001; Blood & Zatorre, 2001; Kampe *et al.*, 2001; Erk *et al.*, 2002; Menon & Levitin, 2005; Izuma *et al.*, 2008; Zink *et al.*, 2008)) but also in conjunction with areas including the insula, putamen and ACC, are involved in emotional responses and reinforcement learning towards aversive stimuli (Knight *et al.*, 2003; Knight *et al.*, 2004; Kalisch *et al.*, 2006; Klucken *et al.*, 2009; Knight *et al.*, 2009; Levita *et al.*, 2009; Liang *et al.*, 2011; Klucken *et al.*, 2012). Activity in the amygdala also links to arousal levels (Anderson & Sobel, 2003; Small *et al.*, 2003; Seeley *et al.*, 2007b), hedonic valence (Kober *et al.*, 2008) and in the context of positive reinforcement processing likely encodes the emotional relevance of a stimulus (Metereau & Dreher, 2013; Sescousse *et al.*, 2013). Additionally, in conjunction with the dACC and fronto-insula cortex, the amygdala forms circuitry involved in a salience processing network, a network that is particularly vulnerable in FTD (Seeley *et al.*, 2007b). NAcc activity correlates with reinforcement intensity (Blood & Zatorre, 2001), has a strong role in evaluating mis-matched in predicted and received positive reinforcement, and therefore may play a key role in the salience of prediction-error coding rather than hedonic encoding per-se (Metereau & Dreher, 2013). Anatomically, the NAcc is particularly affected early in FTD relative to AD and healthy controls (Halabi *et al.*, 2013; Moller *et al.*, 2014) and one can therefore easily visualise how disruption of processes such as emotional evaluation, arousal and salience assignment could underpin the evolution of aversive behaviour to previously innocuous stimuli observed here.

Atrophy of the right hippocampus in the evolution of both environmental sound aversion and music aversion and relative preservation in the development of musicophilia supports a role for this region in regulation of valence assignment. Recent fMRI meta-analyses have revealed that although the hippocampus is not consistently activated in positive reinforcement processing, it does appear to be consistently activated, along with areas including

the amygdala and NAcc, in music emotion processing (Sescousse *et al.*, 2013; Koelsch, 2014). The hippocampus shows functional connectivity with the hypothalamus and may play a key role in the modulation of stress responses showing decreased volumes in chronic anxiety states (Kalisch *et al.*, 2006). Its involvement may be particularly relevant for the evaluation of aversive stimuli, reflecting the involvement observed here: hippocampal activity has been strongly correlated with unpleasant or fear provoking music and hippocampal changes have been demonstrated in response to chronic emotional stressors such as depression and post-traumatic stress disorder, as well as in association with the loss of the ability to feel love, compassion or empathy. Indeed, healthy individuals who have a decreased tendency to experience tender, positive emotions display decreased hippocampal volumes and decreased fMRI activity in response to usually pleasant musical stimuli (Koelsch, 2014). Further, chronic acoustic stressors (tinnitus) lead to both structural and functional changes within the hippocampus (Mahoney *et al.*, 2011; Kraus & Canlon, 2012). Therefore, hippocampal dysfunction may partially underpin the aversive perception of usually innocuous stimuli observed here. Conversely, subjects with parhippocampal and hippocampal lesions have been shown to find usually unpleasant dissonant music abnormally pleasant (Gosselin *et al.*, 2006), mirroring the involvement observed here in the patients who had developed musicophilia.

Healthy adults who display normal hedonic responses to secondary positive reinforcers (money) but impaired reinforcement responses to music (musical anhedonics) have been described (Mas-Herrero *et al.*, 2014) fitting the concept of separable reinforcement processing pathways between reinforcement types. The findings here of dissociation from environmental sound and music reinforcement from primary reinforcement, and of subjects who developed musicophilia in association with the evolution of an environmental sound aversion, support this notion. However, the neuroanatomical substrate described here overlapped irrespective of positive reinforcer type and did not shed further light on this question. Music has been shown to evoke strong autonomic responses, the strength of which correlate with the reported hedonic

effects of the music upon the listener and strength of activity in reinforcement learning regions on fMRI (Blood & Zatorre, 2001; Zatorre & Salimpoor, 2013), and an obvious next step for this work would be examine the physiological correlations of these distorted behavioural responses. Additionally, a functional imaging approach would allow a clearer examination of the extent of overlap or dissociation of the positive reinforcer networks involved by stimulus type and elucidation of further areas not revealed with VBM analysis techniques.

The anatomical correlations in the right superior temporal lobe with any change in hedonic response or the presence of environmental sound aversion, were significant at $p<0.05_{FWE}$ corrected over the whole brain volume. However, the additional regions discovered were only significant after separate small volume corrections, raising the possibility of false positive discoveries. Using a single combined small volume, the additional areas reported in table 7.4 did not reach significance at the prescribed threshold of $p<0.05$. However, using more generous thresholds as recently used in the FTD literature elsewhere (Perry et al., 2015), these associations re-emerged. Therefore interpretation of the VBM findings here require some caution. Repeating this study with a larger sample size would provide greater statistical robustness and allow for confirmation of the findings presented here.

7.6 Chapter conclusions:

Here I have demonstrated that abnormalities of hedonic evaluation of music and environmental sound occur in FTD and AD, with evolution of both abnormally increased and decreased pleasure in the former, and aversion in the latter, symptoms in all groups dissociating from those suggestive of primary positive reinforcement processing deficits. Anatomical correlates were found in key regions implicated in reinforcemwnt processing circuitry and additionally, the right anterior temporal lobe, likely involved in appraisal, adding to the evidence

presented in earlier chapters that symptoms common to FTD and AD reflect distortions of sensory stimulus appraisal, valuation assignment and contextualisation.

Chapter 8: General Conclusions

8.1 Summary

The general aims of this thesis were to explore symptoms within the FTD spectrum that, although not canonical for the division of the three main syndromes, may speak to abnormalities in core physiological processing systems, in particular, the disruption of the normal ability to assign degree of salience to sensory information. Experiments 1 and 5 probed underlying anatomical substrates of little explored symptoms, and experiments 2, 3 and 4 used a variety of salience cues to probe whether physiological responses may be differentially affected between FTD syndromes; the general hypotheses were that as different FTD syndromes bear the brunt of disease burden in overlapping, but seperable anatomical regions, autonomic profiles in response to salient information would differ between groups, potentially providing a starting point for the development of in-vivo dynamic biomarkers. The hypotheses and results for each experiment are summarised below:

8.1.1 Experiment 1: symptoms suggestive of altered sensory perceptions in FTD and AD reflect abnormalities of core sensory coding pathways.

In this experiment behaviours suggestive of abnormal pain and temperature perception were investigated through a semi-structured questionnaire and VBM analysis was used to seek anatomical correlates.

As hypothesised, in patients with FTD syndromes, behaviours suggestive of deficits in sensory processing were common, and rather than simply reflecting a generalized disordered

behavioural system, were underpinned by selective atrophy to a network of areas usually implicated in the sensory signal coding of pain and temperature sensation in the healthy adult. In patients with SD and bvFTD these symptoms were more frequent, with a tendency towards exaggerated behavioural responses in the former and depressed responses in the latter. Key areas included those implicated in the topographic and modality specific encoding of sensory afferent information, homeostasis and awareness of one's own internal bodily state (Craig, 2009) as well as sensory appraisal and attribution of the appropriate degree of salience. These findings are in keeping with those symptoms suggestive of abnormal sensory perceptions and disintegration of the integrity of the processing self-other boundaries and distortions of body schema described in FTD, in particular in conjunction with C9orf72 genetic mutations (Chan *et al.*, 2009; Downey *et al.*, 2012b; Downey *et al.*, 2014; Landqvist Waldo *et al.*, 2014) and may speak to wider underlying processing deficits in sensory encoding. This study provides evidence for the presence of disrupted sensory coding in FTD and characterises symptomatology upon which further physiological studies could be developed.

8.1.2 Experiment 2: Salience encoding from primitive cues is disrupted in FTD and AD

Salience information is potentially carried by an array of low-level sensory cues and here I hypothesised that physiological reactions to approaching sounds, as one such cue, would be abnormal in FTD. In particular the usually greater autonomic responses evoked by perceptually approaching stimuli, which may be an amygdala mediated 'warning cue', would be particularly disrupted in SD, with subjects demonstrating a lack of normal differential response relative to both other healthy controls and potentially other patient groups. Due to the predominantly posterior disease burden in PNFA relative to other FTD sub-types, by contrast, if this effect is indeed mesial temporal lobe driven, then the responses should remain normal in this group. In contrast to these hypotheses, the differential pupil response

was in fact *exaggerated* in SD relative to healthy controls and significantly depressed in patients with PNFA and AD relative both to the healthy control and SD groups. This suggests that auditory salience coding from perceived motion direction is more posteriorly encoded. This study provides evidence that perceived motion direction can be used to differentiate FTD patient groups, and may provide a starting point for development of novel physiological biomarker of these diseases using other low-level auditory cues.

8.1.3 Experiment 3: Salience processing from more complex emotional cues is disrupted in FTD and AD

This experiment aimed to address whether by using pupillometry to index autonomic function, impairment in the normally greater physiological response to highly emotionally valent stimuli could be demonstrated in FTD and AD. The hypothesis was that the behavioural variant form of FTD, with greatest damage to fronto-insular regions would show overall depressed autonomic responses to auditory stimuli, and that bvFTD along with SD, clinically demonstrating the greatest emotional recognition deficits, would display the greatest impairment in physiological reactivity as modulated by emotional valence. In contrast, with clinically little emotion processing deficit, both PNFA and AD would display a normal relationship between pupillary response and valence of sound. Further, I predicted that bvFTD, having the greatest damage to the fronto-insular region, would demonstrate the most deranged physiological reactions and that in SD deranged responses would potentially reflect a damaged semantic system rather than impaired core autonomic reactivity.

In contrast to the hypothesis of overall maximally depressed autonomic responses in bvFTD, depressed responses were seen in all FTD groups, and were preserved in AD relative to healthy older controls. This result is surprising for the PNFA group and possible explanations for this are discussed below. As predicted, the coupling between affective behavioural response

(valence rating) and pupillary response was preserved in patients with PNFA but deranged in bvFTD and SD syndromes. Slightly surprisingly, responses were also deranged in AD. In SD the relationship between valence of sound and subjective rating when measured against the group's ratings rather than the normal pleasantness of a sound as determined by the healthy controls, was present, supporting the hypothesis that derangement in this group may at least partially reflect impaired semantic processing. This study shows that physiological emotional reactivity is disrupted in FTD and AD and provide potential for the evolution of new physiological biomarkers.

8.1.4 Experiment 4: salience processing from semantic evaluation is disrupted in SD

Here the potential salience of meaningfulness, and whether this can be evaluated with autonomic metrics, was investigated. The hypothesis was that usually meaningful sounds would be naturally more salient and evoke greater pupillary responses than acoustically matched meaningless counterparts, and that patients with impaired semantic function would demonstrate a loss of this normal differential response. In contrast to the hypothesis, in the SD group, the group with the greatest non-verbal semantic deficit as measured on the semantic matching task, the normal preferential response to meaningful sounds relative to the acoustically matched but meaningless counterparts was *exaggerated* and could differentiate the SD group from the healthy older controls. Further, in all groups demonstrating an auditory sematic deficit, the magnitude of the differential response to the two sound conditions correlated with degree of semantic impairment. This study demonstrates that the semantic content of environmental sounds is a strong salience cue and that physiological responses can be quantitatively correlated with degree of auditory semantic impairment. This provides evidence that physiological metrics have the potential to provide real-time in vivo biomarkers of the auditory semantic system, both to separate FTD sub-divisions and track disease progression longitudinally.

8.1.5 Experiment 5: Abnormal pleasure responses to environmental sounds and music are underpinned by disrupted reinforcement learning in FTD and AD

Here I investigated the characteristics and underlying neuroanatomical correlations of the evolution of musicophilia, musical aversion and a generalised environmental sound aversion in relation to changes in primary reinforcement processing, namely the evolution of a sweet-tooth. I hypothesised that changes in sound pleasure behaviours would be relatively common, could dissociate both by sound type and by reinforcer type, and that they would be underpinned by damage to brain regions involved in normal reinforcement learning. Symptoms were found to occur commonly in both FTLD and AD, although the prevalence and directionality of the changes differed between groups. Further, symptoms suggestive of abnormal hedonic processing were shown to dissociate by modality with increased pleasure for music occurring in association with decreased pleasure from environmental sounds. There was a strong correlation between abnormalities in reinforce type (sound and sweet tooth) but in some cases these double dissociated with a decrease in pleasure from sound coupled with an increase pleasure from sweet foods. Neuroanatomically, symptoms were associated with atrophy of areas implicated in reinforcement learning and emotion processing circuitry as well at the right anterior temporal lobe, suggesting a role for deranged contextual evaluation. Together, these findings provide further evidence that symptoms observed in dementia are underpinned by abnormalities to key physiological processing systems.

Drawing the results from these chapters together we can form the following conclusions:

8.2 Exploration of symptoms in FTD can demonstrate disruption to brain anatomical pathways involved in stimulus encoding:

Here, abnormal pain and temperature (experiment 1) and pleasure (experiment 5) perceptions in FTD were demonstrated to be underpinned by damage to specific anatomical regions implicated in the normal processing of sensory stimuli, rather than occurring as part of a generalised dis-regulated behavioural disorder. It is of interest that as well as specific regions implicated in processing of pain and temperature (insula and thalamus) in experiment 1 and in reinforcement learning and emotion processing circuitry (NAcc, Amygdala) in experiment 5, the right anterior temporal lobe was involved in both processes. The right ATL has a role in contextualisation and non-verbal semantic appraisal, and this area warrants further investigation using larger groups (see below).

8.2.1 Autonomic responses are disrupted in FTD and AD:

8.2.1.1 *Overall autonomic reactivity is differentially altered in FTD and AD relative to healthy controls:*

In experiment 2, overall pupillary reactivity was found to be depressed in a C9orf72 genetic group relative to the phenotypically matched but non-genetic bvFTD cases and healthy older controls. In experiment 3, using a larger stimulus set with larger patient cohorts, overall pupillary responses were found to be depressed in all FTD groups but normal in AD relative to healthy older controls. Since autonomic reactivity is predominantly a fronto-insular mediated process, the retention of normal overall pupillary reactivity in AD (irrespective of salience value of the stimuli) was to be expected; the depressed reactivity in the PNFA group was, however, somewhat surprising, given the more predominantly dorsal disease burden in this syndrome but may reflect the anatomical heterogeneity of this syndromic group (Warren *et al.*, 2013b). To test this possibility further, syndromic groups' mean pupillary responses for all trials in all three physiological experiments of this thesis (a total of 4523 data points) were combined and

compared (using statistical methods as outlined in chapter 2 to allow for non-independence of data points due to participants involvement in multiple experiments). Results are shown in figure 8.1; with a larger sampling size overall pupillary responses were normal in the PNFA and AD groups relative to controls. In contrast, responses were depressed in bvFTD and SD groups relative to the control (control vs. bvFTD, $p<0.01$, $z=-2.85$, $\chi^2= 47.19$; control vs. SD, $p<0.0001$, $z=-4.65$, $\chi^2=47.19$) and in all FTD groups relative to AD (AD vs. bvFTD $p< 0.0001$, $z=-4.19$, $\chi^2= 38.91$; AD vs. SD $p<0.0001$ $z=-5.7$, $\chi^2= 33.13$; AD. vs. PNFA, $p<0.05$ $z=-2$, $\chi^2= 4.02$) and were further depressed relative to the PNFA group in bvFTD and SD (PNFA vs. bvFTD, 0.05 $z=-1.96$, $\chi^2= 38.91$; PNFA vs. SD <0.0001 $z=3.55$, $\chi^2= 33.1$).

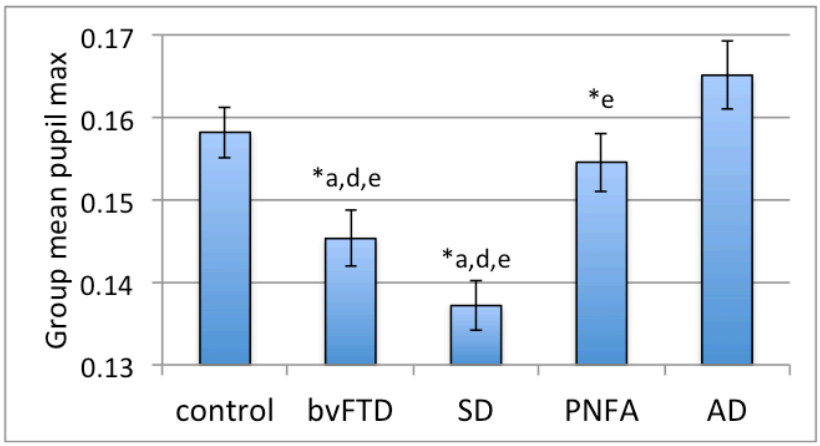

Figure 8.1. pupillary responses ($Pupil_{max}$) for each group averaged over the three pupillary experiments. Bars indicate one standard error. Depressed responses relative to: controls, a; bvFTD, b; SD, c; PNFA, d; AD, e.

Whilst these results do not provide a definitive explanation for the syndrome specific effects of neurodegeneration upon autonomic reactivity, they do support previous work demonstrating depressed autonomic reactivity in FTD (Sturm *et al.*, 2008; Ahmed *et al.*, 2014a). The differences in the findings here and previous work likely reflect the effects of differing sample sizes (as suggested by the refinement of findings as the sample sizes were increased within this work), and use of different stimuli and autonomic metrics (Sturm *et al.*, 2008;

Kumfor & Piguet, 2012; Ahmed *et al.*, 2014a). That phenotypically different syndromes show differentiable physiological signatures provides a starting point for physiologically quantifying these syndromes and conversely physiological signatures may unite patients on more core processing grounds that traverse current syndromic boundaries. That in experiment 2, the genetic syndrome C9orf72 was shown to be differentiable from both phenotypically matched sporadic bvFTD cases and healthy controls has implications not only for differentiating physiological processing stages that ultimately lead to common anatomical (and therefore phenotypical) pathways, but also for early disease detection and tracking of changes; longitudinal study from pre-symptomatic gene carriers would allow evaluation of the sensitivity and specificity of detection of disease onset by physiological reaction monitoring relative to conventional assessment techniques.

8.2.1.2 Salience encoding is disrupted in FTD and AD at several levels of salience processing:

Here I have shown that in healthy older controls, greater pupillary reactions are evoked by sounds that are perceived as approaching, highly valent, and highly meaningful (albeit as a trend in the latter). In both FTD and AD groups these responses are disrupted. In all groups the differences in response towards meaningful vs. meaningless sounds was exaggerated, with in PNFA and AD the normal preferential responses towards motion and emotion, lost, in bvFTD and AD the normal greater response to highly valent sounds lost, and in SD the responses towards motion and meaning exaggerated. Figure 8.2 provides a schematic for a proposed model of how sensory stimulus processing may occur and how this may become disrupted in FTD and AD leading to the clinical pictures observed. Experiments are listed in numerical order from top to bottom correlating to the processing stage they may primarily probe;

Experiment 1 examined the initial brain encoding of afferent sensory information (pale grey top box), whilst Experiments 2-5 examined the assignment of relative salience and positive reinforcer to these stimuli (medium grey middle box). These stages then follow with an effector mechanism for evoking a physiological response (dark grey bottom box). The finding of the correlation of superior colliculus atrophy, a key relay in brainstem mediated pupil response generation, with overall pupillary response in response to salience cues in Experiment 4, is consistent with its proposed role in autonomic effector mechanisms in response to salient cues (Wang *et al.*, 2012; Wang & Munoz, 2014). Those experiments that provided direct anatomical correlations are coded in green with arrows linking to correlated brain substrates, whilst those where no anatomical correlations were found are displayed in purple and linked to putative candidate brain substrates with dashed arrows. Light blue circles indicate those region discovered to be involved and the large grey shaded boxes indicate key networks to which these regions likely belong based upon previous work.

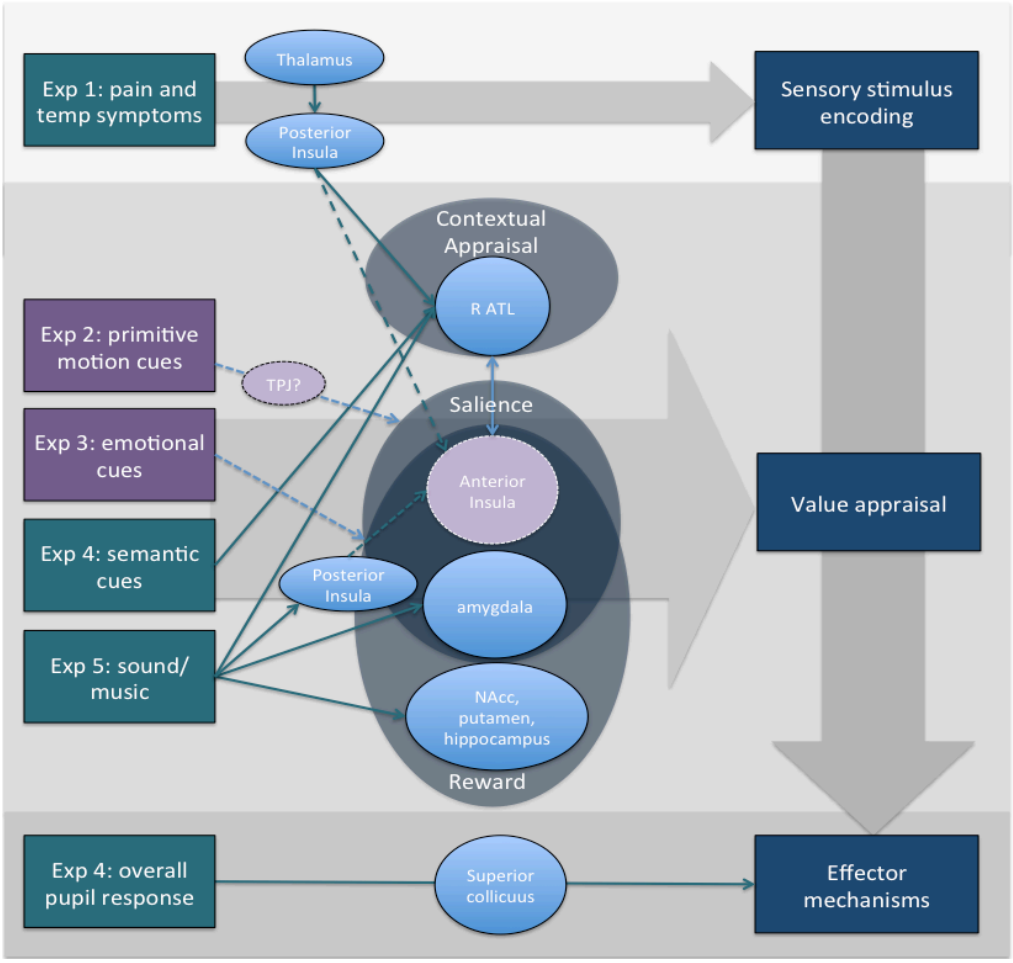

Figure 8.2. Illustration of how the various experiments (left hand boxes) of this thesis putatively probe different stages of an anatomical processing hierarchy (right hand boxes) involved in the physiological encoding of sensory afferent information, to the modulation of this by the relative salience and positive reinforcer value in order to generate an effector physiological outcome. Experiments where anatomical correlations were found are shaded in green with arrows to the regions discovered (blue elipses), and their likely network involvements (dark grey elipses). Those experiments where no direct anatomical correlations were found are contained within purple boxes and are connected to putative responsible anatomical regions with dashed arrows.

8.3. Limitations and Future directions:

8.3.1 Symptom exploration of Experiments 1 and 5:

Experiments 1 and 5 used the results of semi-structured questionnaires to probe symptoms in FTD in more detail and VBM to discover anatomical correlates. The use of retrospectively obtained data from questionnaires is inherently limited for several reasons: firstly, descriptions of particular symptoms cannot be explored in more detail, and two carers may describe the same phenomenon in different language or equally may use similar language to be describing different phenomenon; recording is therefore always inherently going to be biased by the interpretation of the researcher; secondly, some symptoms are intrinsically less accessible to reporting, for example non-painful thermal touch with regards to pain processing, and care givers may be more likely to report patients' behaviour or verbal output where symptoms are heightened rather than attenuated, potentially resulting in bias in the perceived prevalence of different symptom type. Future work should use prospective study of these symptoms with telephone or face-to-face interviews of carers and patients themselves to provide more detail. The symptom constellations exposed here would provide a good starting point for such more details evaluation and as well as using these results as a starting point to prospectively further characterize the breadth and specificity of these symptoms, future work should extend investigation into the physiological domain. A physiological approach (for example measuring behavioural and pupillary correlates to perceived thermal thresholds, or allodynia and dysasthesis to usually innocuous sensory stimuli) could allow direct quantifiable objective markers of abnormalities in pain and temperature processing, with obvious implication for the diagnosis and characterisation of diseases. Further, this line of investigation could be extended to the evaluation of other homeostatic signals that behavioural work suggests may be distorted in FTD, for example changes in eating, sleep or body schema processing and awareness of self in space (Bathgate *et al.*, 2001; Downey *et al.*, 2012a; Downey *et al.*, 2012b; Ahmed *et al.*, 2014b; Downey *et al.*, 2014; Pistacchi *et al.*, 2014). This would also apply to exploring physiological correlates of abnormal reinforcement learning. Deranged

physiological responses to music and sounds would be easy to evaluate. Abnormal pleasure responses to foods (changes in type, quantity, reduced satiety etc) would all be amenable to physiological exploration allowing quantification of change and measurement of progressions in relation to other disease severity markers.

Additionally, VBM is considered a relatively blunt tool for assessing neuroimaging correlates of phenomena. In these studies neuroanatomical correlations were found on a combined group level for FTD. Use of larger sample sizes would hopefully provide the statistical power to allow anatomical correlation of specific symptoms, for example whether exaggeration or depression of pain and temperature responses had a different underlying anatomical substrates, and different FTD sub-group syndromes carried the same or different anatomical signatures for a given symptom change. For example, in both experiment 1 and 5, the right anterior temporal lobe was prominently involved and I would hypothesise that much of this was driven by the SD group.

The Region of Interest based approach used here also produced certain limitations with the possibility of production of false-positive results. In situations where there is adequate evidence from a variety of previous work to suggest that certain brain regions will not have a functional role in a given task, to include these in a whole brain analysis is reducing statistical power un-necessarily and the use of a region of interest based approach is more appropriate. However, the use of several separate regions of interest does treat each analysis as independent and therefore raises the possibility of false- positives. Again, this issue can be addressed more fully in further work by using larger sample sizes to produce greater statistical power. When using VBM it is important to include group membership as a co-variate of no interest to prevent any findings that are more predominant in one group being falsely attributed to atrophy patterns also more common in that group (but not necessarily directly related). However, this potentially minimises results where the symptom under exploration is a function of the region of atrophy in that group. I suspect this is partially the case in experiment 1 to explain why the anterior insula, anterior cingulate or orbitofrontal regions that might have been anticipated a

priori (Craig, 2002; Zhou & Seeley, 2014), were not shown to be selectively atrophied in association with symptom change. Using functional imaging techniques could potentially overcome these issues. In addition to any direct association with atrophy profile, relevant disease effects are likely to reflect connectivity alterations among network elements that are not captured on VBM. Using diffusion tract imaging techniques would allow more direct evaluation of structurally interconnected networks. Ultimately, as the pathological underpinning of a given clinical syndrome remains heterogeneous, histopathological correlations would be required for definitive mapping of pathological substrate to clinical picture.

8.3.2 Autonomic investigations in experiment 2, 3 and 4.

With regards to the pupillometry experiments, this work provides novel insights into the brain physiological processing mechanisms of sensory encoding in neurodegenerative disease and demonstrates that pupillometry lends itself as a viable metric with which to assess these effects.

However, there are several limitations with the approach used here. Firstly, the over-riding aims of this work were to develop in-vivo biomarkers that could physiologically differentiate disease sub-groups from each other and healthy controls. If this could be reliably produced then tracking could be extended longitudinally. Unfortunately it was not possible here to produce responses that could accurately parse out individuals; this is probably best highlighted in experiment 3 where positive quadratic correlations between pupil response and their valence rating (or the 'normal' valence of that sound as determined by the control group mean response) for each sound could not be produced on an individual level. I suspect that this is mainly an effect of noise, both on the level of pupil recordings and of a degree of insensitivity of the experimental stimuli created. For example, in experiment 3 in healthy older controls, when individual pupil responses were compared to the 'normal' valence of a given sound (that

is the average of the control group valence ratings for that sound), a correlation emerged that was not seen between the individual's pupil response and the individual's rating of that sound. Having established the tolerability of this method of data acquisition in behaviourally challenged patients, future work would benefit from refinement of stimulus sets; in particular slightly longer duration experiments would likely be feasible, allowing for the use of larger stimulus sampling sizes over which to average responses. Secondly, the use of repeated measures of a given stimulus, would allow averaging within a stimulus prior to inclusion in group stimulus analysis. Thirdly, the stimuli could be further refined both by breadth and type of stimulus to maximize likely observed differences between stimulus categories; when examining the effects of valence, for example, there are stimuli categories that are particularly emotive, such as erotic vocalisations, that were not included here and stimuli from gustatory, olfactory and visual domains have all been demonstrated to be highly evocative.

With regards to the patient groups used, sample sizes were small and therefore statistical power was limited. Additionally, all individuals were well established in their clinical disease course and in both the VBM and the pupillometry studies included here a particularly interesting angle remaining to be established is whether differences can be detected in presymptomatic individuals (for example C9orf72 genetic mutation carriers) that can then prospectively track change;, this would be feasible considering that there are studies are all ready in place to evaluate these patients across multi-national centres (Rohrer *et al.*, 2015).

8.4 Chapter Summary:

I have presented work here to investigate the neuroanatomical underpinnings of symptoms suggestive of deranged sensory processing in FTD and AD and have demonstrated the validity of the use of pupillometry in conjunction with sound stimuli to assess physiological correlates of these processes. Together these results suggest that symptoms in FTD may be

underpinned by deranged physiological sensory coding systems and suggest putative mechanisms for how these systems may be arranged, providing novel metrics by which to in-vivo assess in real time the effects of disease upon neuronal systems.

Appendix

9.1: Diagnostic criteria for FTD and AD:

Table 9.1.1: Diagnostic criteria for PPA (Gorno-Tempini *et al.*, 2011)

Diagnostic Criteria for PPA
Inclusion: criteria 1–3 must be answered positively
1. Most prominent clinical feature is difficulty with language
2. These deficits are the principal cause of impaired daily living activities
3. Aphasia should be the most prominent deficit at symptom onset and for the initial phases of the disease
Exclusion: criteria 1–4 must be answered negatively for a PPA diagnosis
1. Pattern of deficits is better accounted for by other non-degenerative nervous system or medical disorders
2. Cognitive disturbance is better accounted for by a psychiatric diagnosis
3. Prominent initial episodic memory, visual memory, and visuoperceptual impairments
4. Prominent, initial behavioural disturbance
Diagnostic Criteria for PNFA
I. Clinical diagnosis of nonfluent/agrammatic variant PPA
At least one of the following core features must be present:
1. Agrammatism in language production
2. Effortful, halting speech with inconsistent speech sound errors and distortions (apraxia of speech)
At least 2 of 3 of the following other features must be present:
1. Impaired comprehension of syntactically complex sentences
2. Spared single-word comprehension
3. Spared object knowledge
II. Imaging-supported nonfluent/agrammatic variant diagnosis
Both of the following criteria must be present:
1. Clinical diagnosis of nonfluent/agrammatic variant PPA
2. Imaging must show one or more of the following results:
a. Predominant left posterior fronto-insular atrophy on MRI or
b. Predominant left posterior fronto-insular hypoperfusion or hypometabolism on SPECT or PET
III. Nonfluent/agrammatic variant PPA with definite pathology
Clinical diagnosis (criterion 1 below) and either criterion 2 or 3 must be present:
1. Clinical diagnosis of nonfluent/agrammatic variant PPA
2. Histopathologic evidence of a specific neurodegenerative pathology (e.g., FTLD-tau, FTLD- TDP, AD, other)
3. Presence of a known pathogenic mutation
Diagnostic Criteria for SD
Clinical diagnosis of semantic variant PPA
Both of the following core features must be present:
1. Impaired confrontation naming
2. Impaired single-word comprehension
At least 3 of the following other diagnostic features must be present:
1. Impaired object knowledge, particularly for low- frequency or low-familiarity items
2. Surface dyslexia or dysgraphia
3. Spared repetition
4. Spared speech production (grammar and motor speech)
II. Imaging-supported semantic variant PPA diagnosis
Both of the following criteria must be present:
1. Clinical diagnosis of semantic variant PPA
2. Imaging must show one or more of the following results:
a. Predominant anterior temporal lobe atrophy
b. Predominant anterior temporal hypoperfusion or hypometabolism on SPECT or PET
III. Semantic variant PPA with definite pathology
Clinical diagnosis (criterion 1 below) and either criterion 2 or 3 must be present:
1. Clinical diagnosis of semantic variant PPA
2. Histopathologic evidence of a specific neurodegenerative pathology (e.g., FTLD-tau, FTLD- TDP, AD, other)
3. Presence of a known pathogenic mutation

Table 9.1.2:Diagnostic criteria for bvFTD (Rascovsky *et al.*, 2007)

Diagnostic Criteria for bvFTD
I. Possible bvFTD
Three of the following behavioural/cognitive symptoms (A–F) must be present to meet criteria. Ascertainment requires that symptoms be persistent
A. Early* behavioural disinhibition [one of the following symptoms (A.1–A.3) must be present]:
A.1. Socially inappropriate behaviour
A.2. Loss of manners or decorum
A.3. Impulsive, rash or careless actions
B. Early apathy or inertia [one of the following symptoms (B.1–B.2) must be present]:
B.1. Apathy
B.2. Inertia
C. Early loss of sympathy or empathy [one of the following symptoms (C.1–C.2) must be present]:
C.1. Diminished response to other people's needs and feelings
C.2. Diminished social interest, interrelatedness or personal warmth
D. Early perseverative, stereotyped or compulsive/ritualistic behaviour [one of the following symptoms (D.1–D.3) must be present]:
D.1. Simple repetitive movements
D.2. Complex, compulsive or ritualistic behaviours
D.3. Stereotypy of speech
E. Hyperorality and dietary changes [one of the following symptoms (E.1–E.3) must be present]:
E.1. Altered food preferences
E.2. Binge eating, increased consumption of alcohol or cigarettes
E.3. Oral exploration or consumption of inedible objects
F. Neuropsychological profile: executive/generation deficits with relative sparing of memory and visuospatial functions [all of the following symptoms (F.1–F.3) must be present]:
F.1. Deficits in executive tasks
F.2. Relative sparing of episodic memory F.3. Relative sparing of visuospatial skills
II. Probable bvFTD
All of the following symptoms (A–C) must be present to meet criteria.
A. Meets criteria for possible bvFTD
B. Exhibits significant functional decline (by caregiver report or as evidenced by Clinical Dementia Rating Scale or Functional Activities Questionnaire scores)
C. Imaging results consistent with bvFTD [one of the following (C.1–C.2) must be present]:
C.1. Frontal and/or anterior temporal atrophy on MRI or CT
C.2. Frontal and/or anterior temporal hypoperfusion or hypometabolism on PET or SPECT
III. Behavioural variant FTD with definite FTLD Pathology
Criterion A and either criterion B or C must be present to meet criteria.
A. Meets criteria for possible or probable bvFTD
B. Histopathological evidence of FTLD on biopsy or at post-mortem
C. Presence of a known pathogenic mutation
IV. Exclusionary criteria for bvFTD
Criteria A and B must be answered negatively for any bvFTD diagnosis. Criterion C can be positive for possible bvFTD but must be negative for probable bvFTD
A. Pattern of deficits is better accounted for by other non-degenerative nervous system or medical disorders
B. Behavioural disturbance is better accounted for by a psychiatric diagnosis
C. Biomarkers strongly indicative of Alzheimer's disease or other neurodegenerative process

Table 9.1.3: Diagnostic criteria for AD (Dubois *et al.*, 2007)

Probable AD: A plus one or more supportive features B, C, D, or E
Core diagnostic criteria
A. Presence of an early and significant episodic memory impairment that includes the following features:
1. Gradual and progressive change in memory function reported by patients or informants over more than 6 months
2. Objective evidence of significantly impaired episodic memory on testing: this generally consists of recall deficit that does not improve significantly or does not normalise with cueing or recognition testing and after effective encoding of information has been previously controlled
3. The episodic memory impairment can be isolated or associated with other cognitive changes at the onset of AD or as AD advances
Supportive features
B. Presence of medial temporal lobe atrophy
Volume loss of hippocampi, entorhinal cortex, amygdala evidenced on MRI with qualitative ratings using visual scoring (referenced to well characterised population with age norms) or quantitative volumetry of regions of interest (referenced to well characterised population with age norms)
C. Abnormal cerebrospinal fluid biomarker
Low amyloid β1–42 concentrations, increased total tau concentrations, or increased phospho-tau concentrations, or combinations of the three
Other well validated markers to be discovered in the future
D. Specific pattern on functional neuroimaging with PET
Reduced glucose metabolism in bilateral temporal parietal regions
Other well validated ligands, including those that foreseeably will emerge such as Pittsburg compound B or FDDNP
E. Proven AD autosomal dominant mutation within the immediate family
Exclusion criteria
History
Sudden onset
Early occurrence of the following symptoms: gait disturbances, seizures,
behavioural changes
Clinical features
Focal neurological features including hemiparesis, sensory loss, visual field deficits
Early extrapyramidal signs
Other medical disorders severe enough to account for memory and related symptoms
Non-AD dementia
Major depression
Cerebrovascular disease
Toxic and metabolic abnormalities, all of which may require specific investigations
MRI FLAIR or T2 signal abnormalities in the medial temporal lobe that are consistent with infectious or vascular insults
Criteria for definite AD
AD is considered definite if the following are present:
Both clinical and histopathological (brain biopsy or autopsy) evidence of the disease, as required by the NIA-Reagan criteria for the post-mortem diagnosis of AD; criteria must both be present
Both clinical and genetic evidence (mutation on chromosome 1, 14, or 21) of AD; criteria must both be present